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THE EFFECT OF MATERNAL THYROID FUNCTION ON FETAL THYROID FUNCTION AND DEVELOPMENT*†

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ABSTRACT

Maternal hypothyroidism is not commonly the cause of sporadic fetal hypothyroidism. Seventeen mothers of cretins were examined two months to twenty-four years after the birth of their last cretinous child; of these, 12 were euthyroid, 4 were probably euthyroid and 1 was moderately but definitely hypothyroid. In newborn living pups of dogs thyroidectomized during pregnancy, the serum PBI level, thyroidal I^{131} uptake and thyroid weight in proportion to body weight did not differ from those in pups born of euthyroid dogs.

Maternal hyperthyroidism produced by administration of large doses of hormone may, however, lead to significant transplacental passage of thyroid hormone to the fetus. Two mothers who had each previously given birth to 2 cretins were treated with large doses of desiccated thyroid during their third pregnancies. Each gave birth to a living infant whose clinical appearance, bone maturation, cord-blood PBI and BEI concentrations, and thyroidal I^{131}

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uptakes suggested that an adequate amount of thyroid hormone had been received transplacentally. Subsequent thyroidal I^{131} uptakes after administration of TSH suggested that one of the infants was athyrotic, whereas the other had an apparently normal gland temporarily suppressed by the exogenous hormone. Reasons for believing that the athyrotic infant may have a better prognosis for mental development than cretins born of mothers euthyroid during pregnancy are discussed. Experiments in pregnant dogs fed large amounts of thyroxine also suggest some transplacental transfer of thyroid hormone when the maternal concentration of serum PBI reaches very high levels.

A CRETINOUS child suffers mental retardation presumably because its thyroid gland is unable to supply enough thyroid hormone to promote normal brain development. The ratio of cerebral to total brain weight is higher at the eighth fetal month than at any other time during life, and the total brain weight at birth is normally somewhat over 25 per cent of the final adult total brain weight (1). We (2) have confirmed the findings of Smith and associates (3) that the earlier in life the diagnosis of cretinism is made and treatment with desiccated thyroid started, the higher the eventual intelligence attained. Unfortunately, the cretinous newborn infant apparently has already suffered from lack of thyroid hormone before birth, as manifested by retarded bone growth (4, 5). It is likely, therefore, that if retarded growth and skeletal maturation is to be prevented, an adequate amount of thyroid hormone should be supplied to the child while still *in utero*. Grumbach and Werner (6) concluded from their studies in humans subjects with I^{131} labeled thyroxine and triiodothyronine that "the human placenta is capable of transmitting significant amounts of thyroid hormone but the rate of transfer across this 'barrier' is slow. Therefore, the quantity of hormone transferred from the mother may be insufficient to support normal skeletal and brain development when the fetal contribution is deficient because of a primary thyroidal defect." This excellent work and the work of Postel in guinea pigs (7) measured the rate of transplacental transfer. We have been unable to find reports of work directly measuring net transplacental transfer of thyroid hormone in the dog or human when the amount of circulating thyroid hormone in the mother has been raised to hyperthyroid levels.

We wish to report work in human subjects and in dogs bearing on the questions: 1) Is maternal hypothyroidism the cause of fetal hypothyroidism? 2) Can sufficient net placental transfer of thyroid hormone from mother to fetus be produced to insure an adequate supply of thyroid hormone for normal brain and skeletal development to a fetus with primary hypothyroidism?

METHODS

Seventeen women were surveyed who had previously given birth to at least 1 cretinous child. The diagnosis of cretinism was proved in each child by clinical and laboratory

examinations at the University Hospital as part of a larger survey of cretins in the State of Michigan (2). One-hour, 2-hour, 5-hour and 24-hour thyroidal I^{131} uptakes (8), basal metabolic rates, and serum PBI (9), cholesterol (10, 11), and PBI^{131} (12) determinations were performed (Table 1) as tests of thyroidal function. During a period of at least four, and usually six weeks preceding studies, none of the women received desiccated thyroid or other preparations containing large amounts of iodine.

Fourteen pregnant mongrel dogs were also studied. The dogs were maintained on a diet of "Rival" or "Pard" dog food supplemented by vitamins and approximately 1 gram of sodium chloride per dog daily. In most instances passive immunization was given against distemper. Sodium-*l*-thyroxine was given orally, the total daily amount being administered in fractional doses morning and evening. We were usually successful in avoiding hypoparathyroidism as a complication of total surgical thyroidectomy; persistent hypoparathyroidism developed in only 1 dog. This complication was relieved by feeding supplementary calcium gluconate and concentrated vitamin D preparations. All other dogs received supplementary calcium and concentrated vitamin D preparations for only one or two days preoperatively and postoperatively, or not at all. In a few instances small subnormal concentrations of I^{131} were demonstrated in the thyroid region of the mother dog at delivery, indicating that a small thyroid remnant was left postoperatively. In Table 3 are listed the doses of I^{131} given twenty-four hours before delivery to the mother dogs in order to assay relative I^{131} concentrations in maternal and fetal tissues. Tissues were placed in tared test tubes containing appropriate amounts of Bouin's solution, and their radioactivity was assayed in a well-type scintillation counter, with a pulse-height selector, having a sensitivity of approximately 4.7×10^5 counts/minute/microcurie and a background of 10 to 30 c.p.m. Brain tissue was usually preserved in formalin rather than Bouin's solution. Histologic studies of fixed tissues were carried out by the usual methods. It is the intention to include most of the results of these tissue studies in a subsequent publication. Blood of spontaneously born pups was obtained (for PBI determinations) from the abdominal aorta, using ether anesthesia. Blood from pups born by cesarean section was obtained by similar arterial puncture, cardiac puncture, or directly from the umbilical artery. In order to obtain PBI values for a litter as a whole, the determinations were performed whenever possible on pooled serum, representing approximately equal contributions from the several pups. However, as we found little individual variation among living newborn pups within a single litter, the blood of 1 or 2 pups was taken as representative of the entire living litter, when necessary. Since much greater variation was found in the thyroidal I^{131} uptakes among the members of a single litter, even after considerable attention to prompt handling and blotting of tissues and other technical details, values for thyroidal uptakes in a small fraction of the litter were never accepted as representative of the entire group. Intravenous pentobarbital anesthesia was used for cesarean section. All spontaneously born pups on which data are included in this report were studied within twenty-four hours after birth, unless specifically stated otherwise. Serum PBI and BEI (13) determinations in dogs were carried out by the same methods as in human subjects.

RESULTS

The thyroid function in mothers of sporadic cretins is shown in Table 1. Twelve mothers were clearly euthyroid, 4 (A.L., A.M., A.S. and B.L.) were probably euthyroid, and 1 (S.H.) was hypothyroid. S.H. had received 1–1½ grains of thyroid daily fairly regularly throughout both of her previous pregnancies, and both children were cretins.

TABLE 1. THYROID FUNCTION IN 17 MOTHERS OF CRETINS

Patient	Age (yrs.)	Years since birth of cretin	Clinical status	Thyroidal I^{131} uptake (% admin'd dose)				BMR (%)	Serum PBI (μg./100 ml.)	Serum cholesterol (mg./100 ml.)	Serum PBI^{131} (% admin'd dose per liter at 24 hrs.)
				1 hr.	2 hrs.	5 hrs.	24 hrs.				
P.P.	38	13	Euthyroid	—	11.0	13.0	42	+13	4.5	221	0.031
V.H.	34	8	Euthyroid	—	9.5	15.0	40	−14	7.0	189	0.028
L.K.	51	22	Euthyroid	—	8.9	11.0	26	+10	7.5	240	0.071
P.W.	39	11	Euthyroid	—	9.3	16.0	26	− 7	5.2	186	—
M.A.	53	24	Euthyroid	5.0	7.8	14.0	28	− 7	7.9	251	—
O.L.	29	8	Euthyroid	6.5	9.0	13.0	26	+ 4	—	—	—
B.N.	52	13	Euthyroid	—	3.6	9.0	26	+ 9	—	—	—
H.Z.	37	6	Euthyroid	3.2	4.8	7.8	16	+ 9	—	—	—
G.A.	39	8	Euthyroid	0.9	2.7	9.0	29	− 9	—	—	—
L.B.	33	6	Euthyroid	9.4	13.0	22.0	38	+ 1	—	—	—
V.K.	28	9	Euthyroid	4.2	7.2	13.0	39	− 2	—	—	—
N.P.	32	2/12*	Euthyroid	10.0	12.0	15.0	22	—	—	183	—
A.L.	31	9	Euthyroid	—	10.0	15.0	28	− 3	9.8†	222	0.049
A.M.	42	15	Euthyroid	4.3	4.7	8.8	16	−23	6.6	148	—
A.S.	48	24	Euthyroid	4.4	5.7	8.5	21	−11	3.1	275	—
B.L.	23	10/12*	Euthyroid	—	—	—	—	—	8.4‡	—	—
S.H.	26	2*	Hypothyroid	2.3	2.3	3.6	0.5	−32	—	—	—

* Cretins born in 2 pregnancies. The age of the youngest cretin is given.

† Serum inorganic iodide concentration, 1.6 μg./100 ml.

‡ Twelfth week of pregnancy.

The effect of maternal hypothyroidism on the fetus in dogs is shown in Table 2. Hypothyroid pregnant dogs (Nos. 5 through 9) may be compared with euthyroid pregnant dogs (Nos. 1 through 4). Although the decrease in serum PBI concentration in the thyroidectomized dogs was not striking,

TABLE 2. EFFECT OF CANINE MATERNAL HYPOTHYROIDISM AND HYPERTHYROIDISM ON THYROID FUNCTION OF NEWBORN PUPS

Dog No.	Wt. at full term (Kg.)	Treatment	Method of delivery (C=Cesarean; S=Spontaneous)	Pregnant dog: Serum PBI (μg./100 ml.) before Rx	Pregnant dog: Serum PBI (μg./100 ml.) at delivery	Newborn pups: Serum PBI (μg./100 ml.)	Newborn pups: Thyroid:muscle I^{131} conc. ratio	Newborn pups: Mean wt. of thyroid (mg.) / Mean wt. of pups (Gm.)
				Euthyroid				
1	20	None	C	—	2.4	—	2250± 1240[1]	0.16
2	16.1	None	C	—	2.2	2.0[1.9][2]	3630± 480	0.19
3	7	None	S	—	2.1	4.8[3]	—	0.21
4	22.4[4]	None	S	—	1.6	2.7	—	—
		Mean for Dogs 1–4			*2.1*	*3.2*	*2940*	*0.19*
				Hypothyroid				
5[5]	16.8	Thyroidectomy 9 days before delivery	C	2.6	1.8	3.3[5]	2350± 810[6]	0.23
6	9.2	Thyroidectomy 19 days before delivery	S	2.3	1.0	3.8[a]	—	0.15[4]
7[7]	12	Thyroidectomy 42 days before delivery	—	3.5	1.5 [1.5][7]	—	—	—
8	20.9	Thyroidectomy 45 days before delivery	C	4.7	1.1 [1.1]	2.3[0.5]	3250± 1220	0.16
9	9.9[8]	Thyroidectomy 50 days before delivery	S	3.8[b]	1.0	5.2	—	0.23
		Mean for Dogs 5–9			*1.3*	*3.7*	*2800*	*0.19*
				Hyperthyroid				
10	23.7	Sod.-*l*-thyroxine, 0.6 mg./day for 15 days; then 3 mg./d. for 12 d.; then 10 mg./d. for 6 d. before delivery	C	2.7	17.3 [9.5]	6.1 [2.7][b-9]	2760± 456	0.17
11	8.4	Sod.-*l*-thyroxine, 4 mg./d. for 10 d. before delivery	C	12.7[c]	25.0[d]	—	433± 289	0.21
12	11.2	Sod.-*l*-thyroxine, 11.6 mg./d. for 15 d. before delivery[10]	S	2.2	30[11]	16.8[c]	—	0.15
13	8.3	Sod.-*l*-thyroxine, 3 mg./d. for 13 d.; then 1.5 mg./d. for 13 d.; then 1 mg./d. for 12 d.; then 4 mg./d. for 8 d. before delivery	C	—	25 [25][f]	7.3[3.9][c]	—	0.27
14	5.2	Sod.-*l*-thyroxine, 5 mg./d. for 7 d. before delivery	C	3.8[c]	19.0[15.2]	6.0[4.6][c]	—	0.28
		Mean for Dogs 10–14						*0.22*

[1] Standard error.
[2] Values in brackets represent BEI.
[3] Mean of 4 sera, each from a different pup or pool of 2–3 pups, with values of 4.1, 4.3, 5.2, 5.5.
[4] Postpartum weight. Total weight of pups 2.7 Kg.
[5] Underdeveloped pups not included.
[6] Data on pups dying in first 4 neo-natal days included.
[7] Probable miscarriage. After a greenish vaginal discharge had occurred uterus was found empty at operation for "delivery". Apparent fresh implantation sites were seen grossly. Microscopic study showed deposition of fibrin and leukocytic infiltration at these sites in a hypertrophied uterus with epithelial hyperplasia—compatible with, but not pathognomonic of recent pregnancy.
[8] Weight 3 wks. before term.
[9] Mean of 4 serum pools, each from 3–4 pups, with values of 5.5, 6.0, 6.1 and 6.9.
[10] Mean daily dose; range, 6–20 mg./day. Daily dose for last 4 days before delivery was 12 mg.
[11] The limits of the analytical procedure allow the statement that this value is clearly >25 μg. per 100 ml.; the estimate of 30 is only approximate.
[a] Serum inorganic iodide 6.0 μg./100 ml.
[b] Serum inorganic iodide 2.4 μg./100 ml.
[c] Serum inorganic iodide >10 μg./100 ml.
[d] Serum inorganic iodide 10 μg./100 ml.
[e] Serum inorganic iodide 0.4 μg./100 ml.
[f] Serum inorganic iodide 8.8 μg./100 ml.

it was steady, as indicated by serial weekly PBI determinations (not shown in the table); thus the PBI level at the time of delivery (Table 2) was significantly lower in the hypothyroid than in the euthyroid pregnant dogs ($P < 0.05$). The thyroidal radioiodine uptake, the serum PBI level, and ratio of mean thyroid weight to mean body weight in the living pups from hypothyroid mothers were not significantly different from those of pups from euthyroid mothers.

Dog 6 actually became somewhat myxedematous in appearance. This was the one dog in our series, however, in which overt hypoparathyroidism also developed. Her serum PBI level at term was 1.0 μg. per 100 ml., and at one week after delivery had fallen to 0.5 μg. per 100 ml. Her pups appeared normal at birth, but 5 died within the first four post-natal days. One of the pups of Dog 6 was given 1 μc. of I^{131} seven hours after birth. This pup was deliberately sacrificed twenty-four hours later and is included among the 5 pups who failed to survive post-natally. The thyroid:muscle I^{131} concentration ratio was 1200:1, and the 24-hour thyroidal uptake was 5.7 per cent. These data cannot be compared directly with those obtained in pups whose thyroidal I^{131} uptake had taken place *in utero*, but other preliminary data obtained in our laboratory suggest that the foregoing uptake is not low for a pup. It was clear that Dog 6 neglected her newborn pups. We, therefore, weaned the pups on the third day of post-natal life. Two survived but grew subnormally in subsequent weeks, even after they were able to eat solid food from a dish. Despite our careful attention to prophylaxis and treatment of infections and worm infestations, 1 died at eighty-seven days, weighing only 560 grams. Autopsy revealed no gross abnormality and the thyroid gland was present. The other pup weighed 1250 grams on the one hundred and eighth day and appeared very weak. She was, therefore, given 1 μc. of I^{131} orally and sacrificed on the following day. The thyroid:muscle I^{131} concentration ratio was 639:1, and the thyroidal uptake, 2.4 per cent. The serum PBI level was 2.0 μg. per 100 ml.

Although Dog 5 was found at cesarean section to have 3 well-developed pups, 5 other pups in the uterus were underdeveloped. The data given in Table 2 were obtained from the 3 well-developed ones. The underdeveloped pups appeared to have died a considerable time before the operation, as they were 8 to 10 cm. in length whereas the living pups were 15 to 16 cm. in length. However, the major organs could be grossly identified in the underdeveloped pups and they did not show significant maceration. The radioactivity of sample tissues from 1 of the underdeveloped pups was counted and no specific thyroidal I^{131} uptake was observed, though the thyroid was clearly identified and dissected.

Dog 9 brought no pups to term. She was exceedingly difficult to handle during the study and received 40 to 50 mg. of chlorpromazine intramus-

cularly about once a week at the time of bleedings for PBI samples. She appeared generally ill, though not jaundiced, during the period of presumed miscarriage and it is possible that a reaction to chlorpromazine contributed to this illness. None of the pups in this study had goiter or umbilical hernia. On histologic examination the thyroids of the pups of Dog 5 showed absence of colloid in the follicles, whereas the thyroids of the pups of Dogs 8 and 9 were entirely normal.

The effect of maternal hyperthyroidism on the fetus is also shown in Table 2. Difficulty in gauging the daily dose of thyroxine required to achieve the desired serum PBI level caused some fluctuation in the amount given each day, but only rarely was it completely omitted for a day or so. Dogs 10, 11, 13 and 14 received a constant daily dose for six, ten, eight and seven days respectively before delivery, and Dog 12 during the last four days before delivery received a dose very close to her average dose for the entire treatment period. It is probable that the serum PBI concentration at delivery was fairly representative of a level maintained for several days preceding delivery.

The serum PBI value of 6.1 μg. per 100 ml. for the litter of Dog 10 appears to differ significantly ($P < .05 > .01$) from the mean obtained by pooling the data on pups from both euthyroid and hypothyroid mothers. However, the thyroid:muscle I^{131} concentration ratio fails to demonstrate that enough thyroid hormone crossed the placenta to depress fetal TSH stimulation of fetal thyroid. The thyroid:muscle I^{131} ratio for the litter of Dog 11 differs significantly ($P < .05$) from the mean of the same ratio in the litters of euthyroid Dogs 1 and 2. The serum PBI level for the litter of Dog 12 is clearly elevated.

Instead of I^{131} in the form of iodide, Dogs 13 and 14 each received 600 μc. of I^{131}-labeled thyroxine (containing 0.4 mg. of thyroxine) intravenously twenty-four hours before cesarean section. Maternal and pooled pup sera were extracted with butanol-sodium thiosulfate solution, the extract evaporated, and the residue redissolved in ethanol ammonia solution and chromatographed with appropriate carriers in butanol-acetic acid-water and butanol-dioxane-ammonia solvent systems. In Dog 13 about 70 per cent, and in Dog 14 about 60 per cent of the total maternal serum activity was in the form of thyroxine; the rest was in the form of iodide. In both experiments the labeled iodide concentration in the fetal serum was almost twice that in the maternal, but the concentration of labeled thyroxine in the fetal serum was only about 5 per cent of that in the maternal serum of Dog 13 and about 17 per cent of that in the maternal serum of Dog 14.

The ratio of mean thyroid weight to mean body weight in the litters of the hyperthyroid dogs did not differ significantly from the same ratio in the litters of euthyroid and hypothyroid dogs. The thyroids of the pups

of Dogs 10, 11, 12 and 13 appeared normal on histologic examination, except for possible increased epithelial pyknosis in the thyroid gland of the pups of Dog 12. The thyroid of each of the pups of Dog 14 contained a histologically benign nodule in which the cells did not form follicles, but no abnormalities were noted in the remainder of the thyroid tissue.

As elevated concentrations of serum inorganic iodide may be associated with false elevations of the PBI level, particularly if extra washings of the precipitate are not performed, serum inorganic iodide concentrations, as well as other data bearing on the validity of the results, are reported for most of the instances of high PBI values listed in Table 2. The elevated serum inorganic iodide concentration in mothers receiving large doses of thyroxine is not surprising and does not render the results invalid; Riggs and associates (14) noted that this may occur in euthyroid women fed large doses of desiccated thyroid. However, the PBI value of 12.7 μg. per 100 ml. in Dog 11, obtained before administration of thyroxine had begun, is probably a fortuitous elevation in view of the elevated level of inorganic iodide. Similarly, the elevated inorganic iodide concentration in the serum of the litter of Dogs 13 and 14 confirms the data obtained with I^{131}, *i.e.*, this litter probably received a considerable amount of iodide transplacentally.

The distribution of radioactivity in the tissue of all groups studied is shown in Table 3. Relatively high concentrations of I^{131} were found in placental tissue.

Application of these results to human pregnancy was first studied in Patient S.H. (Table 1). After completion of the studies described in Section 1, she was given a daily dose of 3 grains of desiccated thyroid. A few months later she again became pregnant. Desiccated thyroid was administered in gradually increasing doses, as shown in Figure 1, which also shows the effects of this treatment on various indices of thyroid function. In the thirty-sixth week of pregnancy she gave birth to a male infant, weighing 4 pounds, 13 ounces. The baby was 17¾ inches in length. He did not appear cretinous and seemed vigorous and in no distress. The circumference of the head was 12¾ inches and the anterior fontanelle was open and soft. No thyroid gland was palpable. Except for acrocyanosis and slight jaundice, the results of physical examination on the second day were not remarkable. The infant's complete blood count and urinalysis findings were within normal limits. No thyroid hormone or vitamin preparation likely to contain significant amounts of iodine was administered to the infant during the first ten post-natal days, and the baby was not put to the breast. He was maintained in an incubator for several days after birth. The concentration of PBI in umbilical cord blood was 10.7 μg. per 100 ml. (inorganic iodide 2.0 μg.), and of BEI, 4.5 μg. per 100 ml. On the fourth post-natal

TABLE 3. CONCENTRATION* OF I^{131} IN TISSUES OF CANINE MOTHERS AT DELIVERY AND THEIR PUPS†

Dog No.	Status	Dose of I^{131} (μc.)	No. of pups	*Maternal*				*Fetal*		
				Thyroid	Muscle	Whole blood	Placenta	Placenta	Whole blood	Muscle†
1	Normal	1,775	6	820	0.8	1.6	3.1 [3]‡	4.7	—	1.5
2	Normal	100	7	2,520	0.4	1.8	5.8 [1]	1.8 [1]	1.1	0.7
5	Thyroidectomized	100	3 living	—	0.4	0.6	15.1 [1]	2.4 [1]	1.7	1.1
			4 underdeveloped	—	—	—	11.0 [1]	4.8 [1]	—	41.2 [1]§
7	Thyroidectomized	100	—	—	2.0	1.6	—	—	—	—
8	Thyroidectomized	100	3	—	0.3	0.7	1.1 [1]	3.3 [1]	1.3	0.7
10	Thyroxine-fed	200	14 living	78	0.4	1.5	1.5 [1]	—	0.5 [13]	0.5 [12]
			1 underdeveloped	—	—	—	—	—	—	54.9
11	Thyroxine-fed	2,000	6	458	0.5	2.1	12.7 [3]	13.6 [3]	12.5 [2]	5.4 [3]

* Counts/min./mg. wet weight of tissue/100 μc. of I^{131} administered. In each instance the I^{131} was given to the mother 24 hours before delivery.

† See Table 2 for I^{131} concentration in fetal thyroid relative to muscle.

‡ When less than the full number of pups was sampled for a given tissue, the number sampled is in brackets.

§ I^{131} concentration in thyroid of this pup was 50.8.

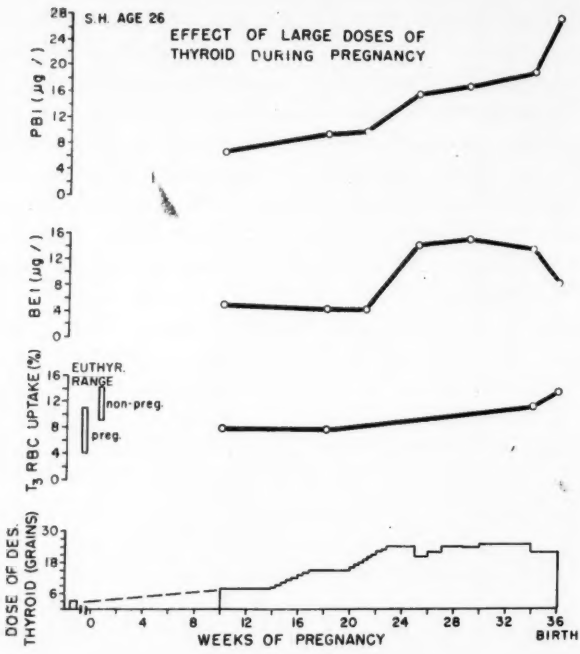

FIG. 1. The effect of desiccated thyroid on the levels of serum PBI and BEI (μg./100 ml.) and on the erythrocyte uptake of triiodothyronine (15) in a pregnant woman, S.H. The patient was taking 3 grains of thyroid daily until the approximate time of conception and then gradually increased the dose at unknown intervals until a dosage of 8 grains daily was reached at ten weeks. The dotted line therefore represents a daily dose of not less than 3 grains and not more than 8 grains at any time. After the tenth week the dose is accurate as shown.

day the infant received 3 microcuries of I^{131} by bottle feeding. The thyroidal I^{131} uptake twenty-four hours later was 6.4 per cent. The infant's serum PBI level on the eighth day after birth had fallen to 3.0 μg. per 100 ml. (inorganic iodide 1.2 μg.). On that day, after the blood for PBI determination had been drawn, he received 10 units of thyroid-stimulating hormone (TSH) (Armour) intramuscularly. On the following day he was again given 3 microcuries of I^{131} by gavage. The two-hour thyroidal I^{131} uptake was 7.4 per cent and the 24-hour uptake was 2.3 per cent. In both instances the infant was carefully observed for a suitable period of time after receiving the I^{131} in order to make certain that he did, in effect, receive and retain the dose intended. A small amount of the first dose was lost, but this was estimated as being clearly less than one quarter of the total intended dose. There was no question that the infant received all

of the second dose; the gavage tubing was carefully rinsed and close observation of the patient thereafter showed that he did not regurgitate any of the dose. A roentgenogram of the infant's legs taken on the fourth day, is shown in Figure 2. At the conclusion of the second uptake experiment, desiccated thyroid was started in a dosage of $\frac{1}{4}$ grain daily. The infant appeared maximally jaundiced on the fourth day; at this time his serum bilirubin level was 14.3 mg. per 100 ml. total (12.6 mg. indirect). The jaundice began to subside promptly, and the bilirubin fell to 9.4 mg. per 100 ml. total (7.5 mg. indirect) by the eighth day. By the eighth day the infant had regained his birth weight. He was able to be taken home by his

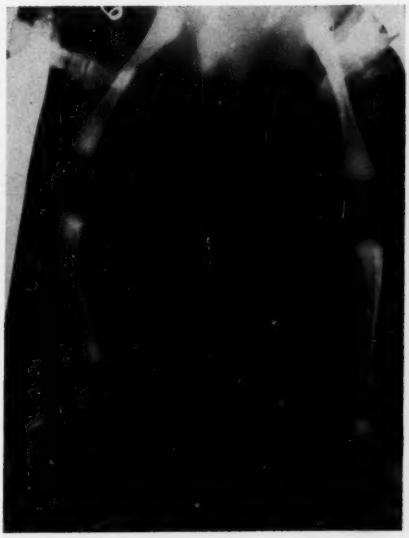

FIG. 2. Roentgenogram of the lower limbs of the infant of Patient S.H. Note the well-developed ossification centers of the distal femoral epiphyses, the absence of the proximal tibial epiphyseal ossification centers and the well-developed talus and calcaneus. The skeletal maturation is therefore normal for 36 weeks. In one of this infant's siblings, roentgenograms of the same areas taken three and a half months post-natally showed virtually no ossification centers in the distal femoral epiphyses.

parents on the twelfth day, at which time he weighed 5 pounds, 3 ounces. He was observed again on the nineteenth day, at which time he weighed 5 pounds, 13 ounces, and was $19\frac{1}{2}$ inches long. He was getting along well, and consuming about 32 ounces of formula-milk in thirty-six hours. Results of physical examination were again essentially normal for a premature infant. The dosage of thyroid was raised to $\frac{1}{2}$ grain daily at this time. A report from the mother indicated that at the age of 6 months the infant was thriving and had shown faster progress in such things as rolling over and babbling than had his siblings at equivalent ages. The dosage of thyroid had been progressively increased. The mother's serum PBI level fell to 6.2 μg. per 100 ml. (BEI 5.8 μg.) on the twelfth day after delivery. At this time her serum cholesterol level was 241 mg. per 100 ml.

Similar treatment was also given to Patient B.L. (Table 1). When she became pregnant a third time, desiccated thyroid was administered as shown in Figure 3. The infant born of this pregnancy was a son who appeared entirely normal. Distal femoral and proximal tibial epiphyseal ossification centers were found to be well developed upon x-ray examination. In the blood of the umbilical cord, the concentration of serum PBI was 8.7 μg. per 100 ml. (inorganic iodide 4.9 μg.), and of BEI, 5.3 μg. per 100 ml. On the fourth post-natal day, a dose of 3 microcuries of I^{131} was given by gavage. The thyroidal I^{131} uptake twenty-four hours later was 2 per cent. On the ninth day the infant's serum PBI level was 8.2 μg. per

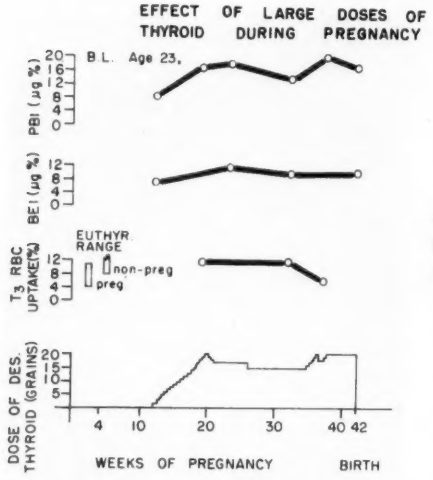

FIG. 3. The effect of desiccated thyroid on the levels of serum PBI and BEI and on the erythrocyte uptake of triiodothyronine (15) in a pregnant woman, B.L.

100 ml. (BEI 6.0 μg.). After blood had been obtained for the foregoing determinations, TSH (10 units) was given intramuscularly. On the following day, a dose of 3 microcuries of I^{131} was again given by gavage. Thyroidal I^{131} uptakes were 7 per cent at two hours and 27 per cent at twenty-four hours. This infant received no desiccated thyroid after birth. When last seen, at 6 weeks of age, he still showed normal progress and there was no suggestion of cretinism.

DISCUSSION

As all but 1 of 17 mothers of cretins were euthyroid, it is unlikely that lack of maternal thyroid hormone is the usual cause of sporadic cretinism. The fact that the mean serum PBI level in pups of hypothyroid dogs was not significantly lower than that in pups of euthyroid dogs supports our observations in human beings. Furthermore, Hodges *et al.* (16) have reviewed the literature and added a case of their own demonstrating that

totally athyroid women can conceive and bear normal children while myxedematous.

Our data suggest also that if the level of circulating thyroid hormone in the human mother is raised to levels greatly above normal by feeding thyroid hormone orally in large doses, the fetal serum PBI level may be raised also.

The low serum inorganic iodide concentration accompanying the elevated PBI concentration in the infant of Patient S.H. is strong evidence against the high PBI value being due inorganic iodide. Furthermore, the low-normal serum BEI level and the normal clinical and x-ray appearance of the infant suggest that an adequate supply of thyroid hormone was present.

In normal infants in the usual weight range, the ossification centers for the distal femoral epiphyses are almost invariably present at birth (17, 18). Yet among 9 cretins at the University Hospital, roentgenograms of the knees taken from one to six months after birth (before treatment) showed that ossification centers for the distal femoral epiphyses were absent in 4 and only faintly visible in 3; moreover, in 1 of the remaining 2 in whom they were visible, the centers were less developed than in the normal newborn child. One of Patient S.H.'s children was among this group of 9; in this infant the ossification centers were only faintly visible at $3\frac{1}{2}$ months of age. In S.H.'s third child, reported here, these ossification centers were present, although the infant was premature and had an inactive thyroid gland.

The evidence that the thyroid of S.H.'s infant was not functioning lies in the decrease in serum PBI concentration during the first post-natal week, at which time it normally rises (19, 20), and the subnormal thyroidal I^{131} uptake after administration of TSH in the absence of an elevated level of serum inorganic iodide.

The normal serum levels of PBI, BEI and inorganic iodide, and the low thyroidal I^{131} uptake before TSH in the infant of Patient B.L. suggest that significant transplacental transfer of thyroid hormone also occurred in this instance. The normal thyroidal uptake after TSH and the subsequent maintenance of normal serum PBI levels in the absence of thyroid therapy suggest that this infant had a normal thyroid which was temporarily depressed in its function by the hormone supplied transplacentally.

At present we have not sufficient data to quantitate the relationship between the maternal level of thyroid hormone and the degree of placental transfer. It is possible that the placenta has a threshold level for maternal serum thyroid hormone concentration below which there is an inadequate net transfer of thyroid hormone from mother to fetus. It is, however, of interest that the ratio of PBI concentration in maternal blood to that in

cord blood in Patient S.H. was 2.5:1 and in Patient B.L., 2:1. This is similar to the maternal:fetal serum PBI^{131} ratio of 2.1:1 found by Grumbach and Werner (6) in their patient who had been given labeled thyroxine eight days before delivery.

Because of differences in placentation between the species, placental permeability cannot be assumed to be the same in man and dog. In the litter of Dog 10, the slight elevation of the serum PBI level and the lack of depression of the thyroidal I^{131} uptake suggest that placental transfer of thyroxine in this dog was either nil or very slight. The data on Dogs 11 and 13, taken together, might be interpreted as indicating more definite passage of thyroxine across the placenta, if it were not for the fact that in the litter of Dog 13 the serum inorganic iodide level was elevated. We unfortunately do not have parallel data for Dog 11. Therefore, the elevated serum PBI concentration may not represent thyroid hormone, for we cannot deny the possibility that pups receiving large amounts of iodide transplacentally may show an elevation of the serum PBI level analogous to that sometimes seen in human subjects who have received large amounts of iodine by mouth. The depression of thyroidal I^{131} uptake might similarly be due to iodide. Even if all the labeled thyroxine in the serum of the litters of Dogs 13 and 14 came from the mother, the rate of transfer was not high in these dogs. The litter of Dog 12 did show elevation of the PBI level without elevation of the inorganic iodide level. Even in this dog we were unable to demonstrate sufficient depression of the fetal pituitaries to depress thyroid weight significantly. As maternal thyroidal I^{131} uptake was depressed by thyroid feeding, it is unlikely that maternal TSH activity significantly influenced fetal thyroid activity.

Objection might be raised to the practice of giving excessive doses of thyroid to a pregnant woman, on the grounds that increased levels of circulating thyroid hormone in the fetus might suppress the development of its own thyroid through depression of fetal TSH activity, and thus create athyroidism in a fetus not otherwise destined to suffer lack of a thyroid. Our follow-up to date suggests that the infant of Patient B.L. has sustained no such ill effects. Although maternal thyrotoxicosis has occasionally been cited as predisposing to cretinism, the evidence for this is slight. One of the cases cited (21) was in fact a cretinous child of an apparently thyrotoxic mother, but this child had a normal twin! Further, Myant's (22) work suggests that human placental transfer of thyroid hormone early in pregnancy is very slight. There is some evidence from work with animals that maternal TSH does not cross the placental barrier (23, 24). In our 2 patients, during the early weeks of pregnancy, we avoided raising the concentration of serum PBI to levels greater than those normally found in pregnancy, in an attempt to allow ample time for fetal

thyroid development. In fact, PBI elevations above normal in Patient S.H. were avoided until after the twenty-first week. This would have allowed ample time for development of the thyroid in the fetus.

The markedly increased serum PBI level in the litter of Dog 12 did not appear to have interfered with thyroid weight or morphology. However, in none of the dogs except No. 13 was a large dose of thyroxine administered to the mother until after fifteen days (about one quarter) of the gestation period had been completed. Administration of thyroxine to Dog 13 began the day after mating.

One might raise another objection to making the pregnant mother artificially hyperthyroid. Astwood (25) and Gardiner-Hill (26) reported that spontaneous thyrotoxicosis in pregnant women is associated with increased fetal loss. However, in spontaneous thyrotoxicosis, more symptoms and signs develop than in euthyroid individuals fed sufficient desiccated thyroid to raise the serum PBI to a comparable level. Our patients, like the nonpregnant women studied by Riggs (14), had relatively few symptoms of thyrotoxicosis.

In older cretins, it has been possible to accelerate skeletal maturation up to three years beyond the chronologic age. One might question whether skeletal maturation could be accelerated *in utero* to the point of premature synostosis of the skull, by means of elevating the thyroid hormone level. Of 9 children reported in the literature (27–32) who were born with thyrotoxicosis, none had definite evidence of accelerated skeletal maturation. Their serum BEI levels were not recorded, however. Our thyroid-deficient child with a normal serum BEI level showed no acceleration of skeletal maturation beyond normal.

We advocate the use of large doses of desiccated thyroid only when there is a greater than normal probability that the pregnant mother will give birth to a cretin; even then, one cannot state the precise maternal serum PBI value that represents the best compromise between the threat of hypothyroidism to the fetus and the possible threat of hyperthyroidism to the mother. We have found, in a survey of 49 cretins (2), that if a woman has given birth to 2 cretins, she has an increased chance of producing a cretin in the next pregnancy. It is also theoretically possible that if a woman has a demonstrable titer of circulating auto-antibodies against the thyroid, these antithyroid antibodies may be transmitted transplacentally to the fetus and damage its thyroid *in utero* (33). We are studying this possibility at present in animals and in human subjects.

It also seems reasonable to expect that administration of large doses of thyroid to the pregnant mother might promote the development of intelligence in the child even when the large doses are started after completion of the first half of pregnancy, as the latter part of pregnancy appears to

be a time of particular importance for the development of the cortex of the brain.

The relatively normal general physical appearance of newborn cretins in contrast to their retarded skeletal development has been noted by many workers. The ability of inadequate doses of thyroid hormone to reverse many of the metabolic and clinical signs of thyroid deficiency while failing to promote optimal skeletal growth and maturation has also been stressed, and it has been pointed out that doses inadequate to promote normal skeletal maturation appear also to be inadequate to promote optimal central nervous system maturation (34). Although it is generally agreed that if skeletal development be retarded at birth, brain development is also retarded (34), skeletal growth may later be brought to normal with doses of desiccated thyroid that still do not induce normal brain development. It is possible that the amount of thyroid hormone crossing the placenta at the serum levels of PBI seen in normal pregnancy may supply a cretinous infant with a definite but inadequate amount of thyroid hormone during the last few months of fetal life, allowing the infant to be born with a fairly normal physical appearance but with the stigmata of the insufficient hormone supply detectable upon x-ray examination of the bones.

We as yet have no proof that the program described here can insure normal brain development of the fetus, but we consider this a fairly reasonable hope.

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THE EFFECT OF IODIDE ON THE ACTION OF THYROTROPIN IN THE HYPOPHYSECTOMIZED PATIENT*†

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ABSTRACT

Six previously euthyroid, hypophysectomized patients were given thyrotropin by injection twice daily to stimulate thyroidal uptake and secretion of radioactive iodine. In the presence of a goitrogen-induced block of organic binding of iodine and during continued administration of thyrotropin, iodide produced a significant decrease in the rate of loss of I^{131} from the thyroid gland in only 1 patient. Propylthiouracil without thyrotropin did not increase the rate of loss of I^{131} from the thyroid of a hypophysectomized patient. Thyrotropin was used in a dosage of 10 units per day in 4 studies and 2 units per day in 3 studies. The rate of loss was less with 2 units than with 10 units, whether or not iodide was administered. Iodide decreased the rate of loss to the same extent in the one patient in whom it was significantly effective, at both levels of thyrotropin.

THYROTROPIN increases the rate of release of iodine from the thyroid in the presence of a methimazole block of organic binding, as measured by the slope of a semi-logarithmic plot of the I^{131} remaining in the gland (1). Goldsmith and co-workers (2) have demonstrated this effect in hyperthyroid patients and Solomon (3) obtained similar results in euthyroid subjects.

The administration of iodide to patients with Graves' disease markedly decreases the rate of release of I^{131} from the thyroid in the presence of a goitrogenic block (4, 5). The injection of thyrotropin can cause acceleration of the release of I^{131} to rates approximately equalling those observed before iodide was given. Greer and co-workers (4, 5) suggested that the action of iodide in ameliorating thyrotoxicosis resulted, in part, from an inhibition of the secretion of thyrotropin by the pituitary.

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In euthyroid subjects, on the other hand, the effect of iodide on the rate of release of radioiodine from the gland is unsettled. In the absence of exogenous thyrotropin, iodide apparently has no effect (5). During injection of thyrotropin, however, both Solomon (3) and Greer (5) observed that iodide decreases the discharge rate in a majority of euthyroid persons.

The present investigation was undertaken to determine whether the administration of iodide can alter the acceleration of the discharge rate produced by thyrotropin in the hypophysectomized human. Hypophysectomized patients are ideal subjects for the study of the effect of iodide on the action of thyrotropin, since endogenous sources of this hormone are excluded. Solomon (3) reported an increase in the net rate of loss of I^{131} from the unblocked thyroid of a patient with panhypopituitarism given doses of thyrotropin ranging between 1 and 4 units per day. Li and coworkers (6) demonstrated a decreased collection of radioiodine by the thyroid and a fall in the serum level of protein-bound iodine (PBI) to myxedema levels within four weeks following hypophysectomy. In the absence of the pituitary, thyrotropin is effective in restoring the uptake to normal (6) and in increasing the secretion from the gland (3, 6).

METHODS AND MATERIAL

Methods

Six patients were studied at various intervals between four weeks to fourteen months after hypophysectomy for metastatic carcinoma.[1] Thyroid function was normal prior to the operation in all patients. These patients were presumed to have had total hypophysectomy, on the basis of lack of urinary gonadotropins and markedly depressed secretion of Porter-Silber chromogens. Before the study was started, the thyroidal I^{131} uptake was low in all patients, and the serum PBI level was in the myxedema range in 4 of the 5 patients in whom it was determined (Table 1).

Thyrotropin[2] was given throughout 4 studies in a dosage of 5 units intramuscularly every twelve hours. In 3 studies an initial single dose of 10 units was given to ensure an adequate uptake of I^{131} and the dose was then reduced to 1 unit every twelve hours for the duration of the study. On the third day of administration each patient received a labeling dose of I^{131} (about 1 millicurie) orally. The radioiodine in the thyroid was measured daily using a collimated scintillation counter with an aliquot of the dose as a standard. Counting times were sufficient to yield a maximum statistical variation of 2 per cent. In 4 studies the serum PBI^{131} level was measured daily throughout the study.

After the 48-hour thyroidal uptake was determined, each patient was given a thyroid blocking agent (100–200 mg. of propylthiouracil or 20 mg. of methimazole every eight hours) for the duration of the study. After 4 to 11 measurements of the thyroid I^{131} uptake to establish the rate of loss, a solution of potassium iodide (1 to 2 Gm. of iodine as KI) was administered by mouth daily in fractional doses for the duration of the study.

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[1] The patients were made available for study through the courtesy of Drs. Olaf H. Pearson and Bronson Ray.

[2] Thyrotropar was kindly provided by Dr. M. C. Wynes, Armour Laboratories, Chicago, Illinois.

The I^{131} in the thyroid was measured for four to fourteen additional days during the administration of thyrotropin, propylthiouracil and iodide.

To make certain that propylthiouracil alone would not alter the rate of loss of I^{131}, a second, modified study was undertaken in 1 patient (L.H.). The thyrotropin was discontinued twenty-four hours after the administration of I^{131}, and the thyroidal I^{131} was measured daily for five days. Following this, propylthiouracil was given in a dose of 150 mg. every eight hours for the duration of the study. After six days of treatment with propylthiouracil, thyrotropin was again administered. Because of an urticarial reaction, only 3 further injections of thyrotropin (5 units each) were given.

In the study of 1 patient (M.Q.) with the larger amount of thyrotropin, the thyroidal I^{131} was measured for eight days after thyrotropin was stopped and while iodide and methimazole were continued.

Patients

A.L. Six weeks before the study, hypophysectomy was performed on this 38-year-old woman for mammary carcinoma metastatic to bone. A preoperative thyroidal I^{131} uptake was 48 per cent in forty-eight hours. Postoperative maintenance therapy consisted of prednisone, 5 mg. every eight hours, and injections of Pitressin tannate in oil as needed.

L.H. Three weeks before the first study, hypophysectomy was performed on this 60-year-old woman for mammary carcinoma metastatic to bone and soft tissue. Preoperatively the serum PBI level was 5.5 micrograms (μg.) per 100 ml. Postoperative maintenance therapy was prednisone, 5 mg. every eight hours. Following the first study, the patient received liothyronine (3:5:3′-*l*-triiodothyronine), 75 micrograms daily, for one year until one week before the second study.

A.F. Six weeks before the study, hypophysectomy was performed on this 67-year-old man with metastatic carcinoma of the prostate. Maintenance therapy consisted of prednisone, 5 mg. every eight hours, and Dilantin, 300 mg. daily. Preoperative studies of thyroid function were not made. Signs of myxedema developed during the time between hypophysectomy and the study, and regressed when thyrotropin was administered.

H.G. Fourteen months before the study, hypophysectomy was performed on this 69-year-old man for carcinoma of the prostate metastatic to the bone. Maintenance therapy consisted of prednisone, 5 mg. every eight hours and liothyronine, 75 micrograms daily. Liothyronine was discontinued one week prior to the study.

C.C. Seven weeks before the study, hypophysectomy was carried out on this 54-year-old woman for carcinoma of the breast metastatic to the bone. She had been previously adrenalectomized and oophorectomized. Before hypophysectomy the serum PBI level was 8.3 μg. per 100 ml. and the thyroidal I^{131} uptake was 28.3 per cent of the dose at twenty-four hours. Maintenance therapy consisted of 9-alpha-fluorohydrocortisone (0.4 mg. daily), cortisone (50 mg. daily), and Pitressin tannate in oil three times a week.

M.Q. Eleven weeks before the study, hypophysectomy was performed on this 67-year-old woman for chest-wall recurrence of breast carcinoma. Preoperatively the serum PBI level was 9.0 micrograms per 100 ml. Postoperative maintenance therapy consisted of cortisone, 75 mg. daily, and posterior pituitary powder by nasal insufflation. The first study was made using the smaller dose of thyrotropin. Twenty-nine days after the labeling dose of I^{131}, iodide was stopped and propylthiouracil and thyrotropin were continued. On the thirty-fourth day, when the serum total iodine level had decreased to within 1 microgram of the serum PBI level, propylthiouracil was discontinued and the dosage of thyrotropin was changed to 5 units every twelve hours. On the thirty-sixth day a second labeling dose of I^{131} was given and the study was repeated with the larger dose of thyro-

tropin. On the third day of this second study, fever appeared and methimazole was substituted for propylthiouracil. On the sixth day chloramphenicol (1 gram daily) and prednisone (5 mg. daily) were added to combat a pulmonary infection. These were continuously administered until the end of the study.

RESULTS

In each study the administration of thyrotropin produced an increase of thyroidal uptake of I^{131} from definitely low values to normal or borderline high values (Table 1). The serum PBI level was abnormally low (normal range, 3.6 to 8.8 μg. per 100 ml.) (7) in all except 1 patient prior to the use of thyrotropin. Seven or eight days of thyrotropin administration were required to produce the maximal elevation of PBI concentration shown in Table 1. In 4 studies the PBI level became elevated above the normal range. The magnitude of change in thyroidal I^{131} uptake and in serum PBI concentration was not different for the 2 dosage schedules of thyrotropin used, possibly because of the single dose of 10 units used at the beginning of the lower-dose study. The rate of loss of I^{131} during administration of thyrotropin was significantly less for the 2-unit dosage than the 10-unit dosage, regardless of whether or not iodides were administered (Table 2).

TABLE 1. EFFECT OF THYROTROPIN ON THYROIDAL I^{131} UPTAKE AND SERUM PBI LEVEL IN HYPOPHYSECTOMIZED PATIENTS

Patient	Dose of TSH (units/day)	Thyroidal I^{131} uptake (% dose at 48 hrs.)		Serum PBI (μg./100 ml.)	
		Before TSH	During TSH	Before TSH	During TSH
A.L.	10	7.0	44.7	—	18.9
L.H. 1)	10	3.4	30.2	1.9	12.4
2)*	10	5.8	27.8	.5	4.2
A.F.	10	9.0	36.3	1.9	7.3
H.G.	2	7.4	39.5	1.1	4.2
C.C.	2	3.8	56.5	5.2	8.9
M.Q. 1)	2	6.6	46.0	2.7	8.8
2)*	10	—	23.2	—	9.0
Average		*6.1*	*38.0*	*2.2*	*9.2*

* See clinical abstract for details of second study on this patient.

Iodide effect at higher TSH dosage

Thyroidal I^{131} decreased at a rapid rate during the administration of propylthiouracil and the 10-unit dosage of thyrotropin. The slope of a semi-logarithmic plot of the I^{131} remaining in the gland represents the rate of loss of iodine from the thyroid (2). Table 2 shows the fractional rate of loss and standard errors (in per cent per day) calculated by the method of Kenney and Keeping (8) for the 4 studies in which a 10-unit dose of thyrotropin was injected daily. Rates and standard errors were checked on an electronic computer with the assistance of Dr. Mones Berman of the Department of Physics, Sloan-Kettering Institute. The rates observed during treatment with only propylthiouracil and thyrotropin approximate the average rate for thyrotoxicosis—7 per cent per day as determined by Solomon (2), or 14.2 per cent per day as reported by Greer and DeGroot (5).

TABLE 2. EFFECT OF IODIDE ON THE RATE OF LOSS OF I^{131} FROM THE THYROID DURING ADMINISTRATION OF THYROTROPIN

Patient	Dose of TSH (units/day)	Rate of loss* (% per day)		Significance of difference P
		Propylthiouracil	Propylthiouracil +Iodide	
A.L.	10	17.5±1.7	13.7±1.5	>.05
L.H., 1st study	10	14.0±1.4	8.6±3.3	>.05
A.F.	10	7.3±2.4	9.0±2.2	>.1
M.Q.,† 2nd study	10	8.3±0.7	2.3±0.7	<.005
H.G.	2	5.6±0.7	4.9±0.7	>.1
C.C.	2	5.1±1.8	1.6±1.6	>.05
M.Q., 1st study	2	5.1±0.7	2.3±0.2	<.005

* Slope and standard error of a least-squares fit in a semi-log plot.

† Methimazole used in place of propylthiouracil.

In Patient A.F., who received the larger dose of thyrotropin, the addition of iodide to the regimen had no detectable effect (Fig. 1). In 2 other patients, iodide produced a decrease in the rate of loss, which was significant at the 10 per cent level but not significant at the 5 per cent level. In Patient M.Q., however, a highly significant decrease in the rate of loss occurred when iodides were administered (Fig. 2).

In order to determine whether or not iodide caused an immediate discharge of I^{131} from the gland, a measurement of the thyroidal I^{131} of Patient A.F. was made thirty minutes after the iodide was first administered. No change was detected (Fig. 1).

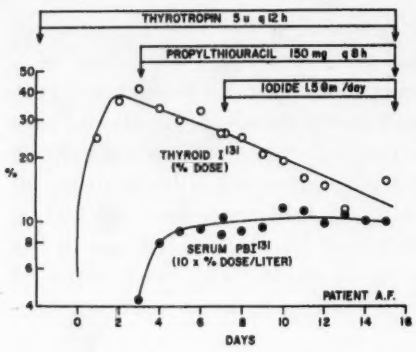

FIG. 1. The effect of iodide on thyroid release rate with 10 units of thyrotropin per twenty-four hours. There was no significant change in the slope.

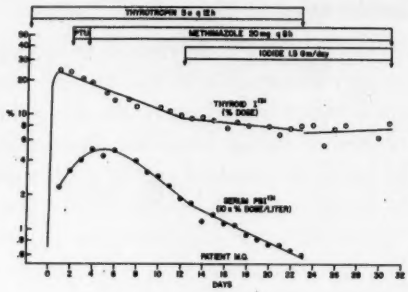

FIG. 2. (Second study on Patient M.Q.). The effect of iodide on thyroid release rate with 10 units of thyrotropin per twenty-four hours. The release rate was significantly decreased by iodide with 10 units of thyrotropin daily. No further release occurred when thyrotropin was stopped.

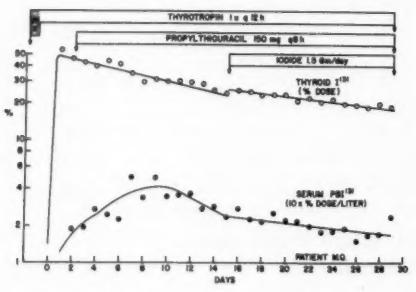

FIG. 3. (First study on Patient M.Q.). The effect of iodide on thyroid release rate with 2 units of thyrotropin per twenty-four hours. The rate was significantly decreased by iodide.

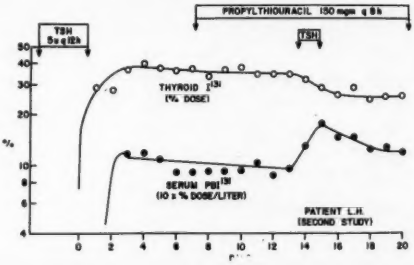

FIG. 4. The effect of propylthiouracil on the release of I^{131} from the thyroid in a hypophysectomized patient. Thyrotropin was used to stimulate uptake of I^{131}. Note the fall in thyroidal I^{131} and the increase in serum PBI^{131} concentration when 3 further injections of thyrotropin were given.

Iodide effect at lower TSH dosage

In studies using the 2-unit dose of thyrotropin daily, the rate of loss of I^{131} from the thyroid was about 5 per cent per day before iodide was administered. This value was less than the rate of loss produced by the 10-unit dose (Table 2). The rate of loss which occurred during the administration of 2 units daily was nearly as low as the rate of 3.5 to 4.9 per cent per day reported from 3 different groups of euthyroid subjects receiving

methimazole (5). It is somewhat higher than the value of 1.9 per cent per day found by Solomon (3).

The addition of iodide to the regimen produced no change in rate of loss for Patient H.G. and a marked decrease in the rate in Patients C.C. and M.Q. (Table 2). The change in Patient C.C. was statistically significant at the 10 per cent level only, however, because of marked variability of the data during the iodide phase of this study. Determinations were made with the patient in her bed at this time because of her poor general condition. The decrease of the rate in Patient M.Q. was statistically significant (Fig. 3).

Modified studies

Propylthiouracil did not alter the rate of loss of I^{131} from the thyroid in a hypophysectomized patient in whom it was used alone after uptake was complete (Fig. 4). The rate of loss of I^{131} from the thyroid of Patient L.H. was 1.2 ± 2.2 per cent per day without propylthiouracil and 0.7 ± 0.9 per cent per day with propylthiouracil. These slopes are not significantly different from zero (no loss). When thyrotropin was discontinued in the last phase of the study on Patient M.Q. during the 10-unit dosage (Fig. 3), the thyroid stopped losing I^{131} even though the iodide and the blocking drug were still being administered.

DISCUSSION

The rate of loss of radioiodine from the thyroid probably represents the rate of secretion of hormonal iodine from the gland. This conclusion is based on the observation (9) that the circulating organic I^{131} in hyperthyroid subjects receiving thyrotropin is indistinguishable from thyroxine and 3:5:3′-triiodothyronine in electrophoresis and paper-strip chromatography.

We have assumed in this study that the propylthiouracil or methimazole prevented the reincorporation of iodine into thyroid hormone in hypophysectomized patients. Greer and Shull (10) reported that doses of 200 mg. of propylthiouracil or 20 mg. of methimazole were occasionally insufficient to prevent the incorporation of I^{131} into gland hormone in euthyroid human subjects. More recently Goldsmith and co-workers (9) demonstrated that 40 mg. of methimazole every six hours prevents any change in the specific activity of circulating precipitable iodine during the administration of 1 mg. of iodide daily to hyperthyroid patients.

In human subjects with intact pituitary glands, Stanbury *et al.* (11) found that the rate of secretion estimated in the presence of a goitrogen-induced block of organic binding of iodine was greater than that estimated by other methods (see Riggs (12)). Our data on 2 patients, however, sug-

gest that goitrogens do not increase the rate of loss in the hypophysectomized patient. In the second study of Patient L.H. (Fig. 4), the rate of loss was unchanged when propylthiouracil was given without the simultaneous administration of thyrotropin. In the second study of Patient M.Q. (Fig. 2), there was no further loss of I^{131} from the gland after the cessation of thyrotropin injections. Thus it is likely that the pituitary is necessary for the increase in the rate of loss induced by the goitrogens.

Iodide decreased significantly the rate of loss of radioiodine from the thyroid in 2 separate studies on the same hypophysectomized patient. This effect of iodide on the action of thyrotropin was of the same order of magnitude as reported by others (3, 5, 9) in non-hypophysectomized patients. These studies would confirm the conclusion that iodide inhibits the action of thyrotropin on the thyroid rather than decreases its secretion by the pituitary. Iodide did not completely inhibit the action of thyrotropin, however, even in doses in excess of those given by other workers.

The other studies reported in this paper were inconclusive regarding the effect of iodide. It must be emphasized that the significance of any differences produced by iodide will depend not only on the magnitude of the difference, but also on the duration of the experiment, the number of measurements, and the accuracy of the measurements. Possibly a significant decrease in discharge rate might have been detected in 3 additional patients had these factors been optimal. Since the first 3 patients studied failed to respond to iodide while receiving 10 units of thyrotropin, it seemed possible that the lack of effect was due to the high dosage of thyrotropin. The experiment with Patient M.Q. casts doubt on this interpretation, however, since iodide in this patient caused a marked decrease in the discharge rate at both the 2-unit and the 10-unit dosage levels of thyrotropin.

If it be assumed that the thyroid gland of the normal human subject is in a steady state with regard to iodide uptake, hormone formation and hormone secretion, then the administration of iodide must produce either hyperthyroidism or a decrease in one or more of the rate constants describing these functions of the gland. In other words, the fraction of one or more of the iodine compartments turned over in a given time must be decreased when iodide is given, or an increase in hormone production will result. The present data, together with the work of others described here, suggest that one of the control mechanisms may be an iodide inhibition of the action of thyrotropin on the gland. From this type of study it is impossible to determine whether iodide acts on the thyrotropin in the blood, at the thyroid gland, or on some intrathyroidal system controlled by thyrotropin. Pituitary mechanisms of control are not excluded in the intact subject by this evidence, but they would seem unnecessary.

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STUDIES ON THE EFFECT OF THYROTROPIN ON HUMAN THYROID FUNCTION*

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ABSTRACT

Time-response and dose-response relationships between a single intramuscular injection of thyroid-stimulating hormone (TSH) and its effect on thyroid function were studied in human subjects without thyroid disease. The effect of TSH on thyroidal accumulation of radioiodine was demonstrable after a latent period of eight to ten hours. It reached its maximum after eighteen to twenty-four hours, decreased to pre-TSH values after five to six days, fell to levels significantly below the pre-TSH values from the eighth to eighteenth days after injection of TSH, and then returned to the original level. The rate of release of thyroid hormone had already started to increase within the first ninety minutes after injection of TSH, and did not return to the pre-TSH rate for at least twenty-four hours. The effect of TSH on the radioiodine-accumulating capacity of the thyroid increased continuously with mounting doses from 0.003 to 0.025 USP unit per Kg. of body weight, but no additional rise was obtained after a further increase of the dose of TSH. The rate of thyroid hormone release, however, also increased continuously with mounting doses of TSH above 0.025 unit per Kg.

THYROTROPIN (TSH) activates all partial functions of the thyroid gland hitherto investigated. In this report we present an investigation, in euthyroid human subjects, of the time-response and dose-response relationships between a single injection of TSH and its effect on the thyroidal accumulation or radioiodine and thyroidal secretion of hormonal iodine. These relations have been studied fairly extensively in different animal species but reports dealing with human subjects (1–3) are still sparse.

MATERIAL AND METHODS

Patients without thyroid disease were selected from a cancer clinic as test subjects. Those in poor general condition and with known heart or renal disease were omitted, as were those who had received drugs known to influence thyroid function.

Thyrotropin ("Thytropar," Armour) was given by intramuscular injection. All thyrotropin used was from one lot (No. R 2506). Carrier-free sterile I^{131} solution was obtained from Amersham, England. I^{132} (half-life 2.3 hours) was distilled at the laboratory from Te^{132} (half-life 77 hours) according to a method described by Cook *et al.* (4).

Thyroidal uptake of I^{131} after twenty-four hours (T_{24}) was measured with a sodium iodide (T1) scintillation counter, provided with a wide-angle lead collimator. The crystal-

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† Director, Professor Sven Hultberg.

to-skin distance was 30 cm. and the influence of secondary radiation was reduced by a 2-mm. lead filter. The standard solution of I^{131} was measured in a fluid volume of 20 ml. contained in a cylindric vessel of 3.7 cm. diameter.

The accumulation rate of radioiodine in the thyroid was studied by means of a method described by Larsson and Jonsson (5, 6) and by Einhorn *et al.* (7, 8). The tracer dose of I^{131} or I^{132} was injected intravenously and the counting rates measured over the thyroid region were continuously recorded by means of a counting rate-meter and a pen writer. Crystal-to-skin distance, filtration, and method for measuring the standard solution of I^{131} or I^{132} were the same as described for the determination of T_{24}. The counter was provided with a 12°-angle conical lead collimator. The difference between the counting rates at eleven minutes and one minute after the injection was expressed as a percentage of the counting rate obtained from a standard solution of I^{131} or I^{132} with the same activity as the tracer dose administered. This value (in the following, called H) was also influenced by changes in extrathyroidal radioiodine within the volume of tissue scanned by the counter. Thus in patients with a very low thyroidal radioiodine uptake, H had negative values due to the fact that the decrease in body background preponderated over the increase in thyroidal radioiodine during the test period. The H value could, however, be used as an arbitrary index for the accumulation rate of radioiodine in the thyroid, and the test had the advantage of great simplicity and short duration. When I^{132} was used, the test could be repeated within relatively short intervals in the same test subject, which was of special value when the time-response relation between injection of TSH and radioiodine accumulation in the thyroid was studied. Fifty microcuries (μc.) of I^{131} or 30 μc. of I^{132} sufficed for determination of the H value. A correction for residual radioactivity in the thyroid was made on the assumption that this activity changed at the same rate during the test period as during the ten-minute period immediately preceding injection of the new tracer dose. As regards I^{132}, a correction was also made for physical decay.

Protein-bound radioiodine in plasma (PBI^{131}) was determined after precipitation of plasma with trichloracetic acid, washing of the precipitate, and measuring of it in a well scintillation counter. Protein-bound stable iodine in serum (PBI) was determined according to Barker *et al.* (9).

As regards T_{24}, H, and serum PBI concentration the results were expressed as the difference between the value obtained after injection of TSH and the value obtained in the same subject before TSH. These differences are, in the following, called T_{24} difference, H difference and PBI difference. As regards PBI^{131}, the results were instead expressed as the percentile change in the pre-TSH value, since the absolute increase of PBI^{131} after TSH was found to be correlated to the pre-TSH value. For determination of the standard error of the mean and the probability value (P), conventional statistical methods were used.

RESULTS

The time-response relationships observed are condensed in Figures 1–3. There were major individual variations in the magnitude of response as measured by the methods employed, though the time curves had a characteristic course in the patients studied.

With the technique and the TSH doses used (0.15 USP unit per Kg. of body weight), no significant changes in the iodine accumulation rate were observed during the first eight hours after the injection of TSH. Ten hours after injection of TSH the accumulation rate of iodine increased in all

patients studied. From the eighth to the eighteenth hour after the injection, all of the tested patients exhibited a continuous rise in the H value. In some patients the effect reached its maximum after eighteen hours and in others not until twenty-four hours after the injection of TSH. After twenty-four hours it gradually decreased and, from eight to eighteen days after the injection, there was a period of subnormal values before the return to the original level (Fig. 1). The increase in the rate of thyroid hormone release started much earlier, as indicated by a significant ($P < 0.001$) elevation of PBI^{131} values observed ninety minutes after the injection of TSH (Fig. 2). During the last interval studied (twenty-four to forty-eight hours) the percentile increase of PBI^{131} per hour was still significantly greater ($P < 0.05$) than before the injection of TSH (Fig. 3). The increased rate of hormone release was thus maintained for more than twenty-four hours after injection of TSH.

The dose-response relationships observed are presented in Figures 4 and 5. TSH doses ranging from 0.0031 to 0.3 USP unit per Kg. of body weight were tested. The H value and 24-hour thyroidal I^{131} uptake rose continuously with increasing doses of TSH from 0.0031 to 0.025 unit per Kg., and no additional rise was observed after a further increase of the dose (Fig. 4).

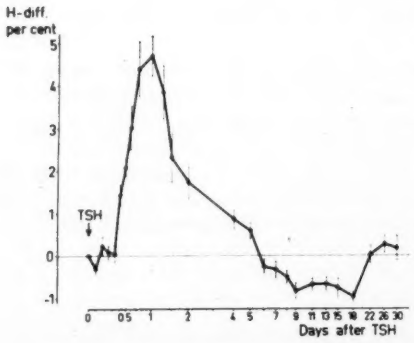


FIG. 1. Time-response relationships after a single intramuscular injection of TSH (0.15 USP unit per Kg. body weight). Duration of the effect on the accumulation rate of radioiodine, and rebound phenomenon. The standard error of the mean is indicated. Each point represents data on 10 to 35 patients.

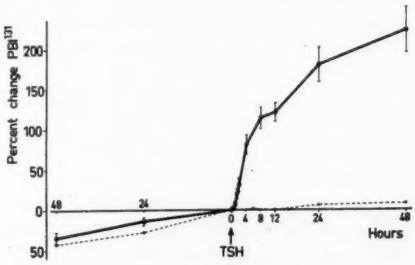


FIG. 2. Time-response relationships after a single intramuscular injection of TSH (0.3 USP unit per Kg. body weight) given four days after a dose of I^{131} (0.3 mc. I^{131}). The effect on PBI^{131} concentration in plasma was significant ($P < 0.01$) ninety minutes after TSH injection. The percentile change has been calculated in relation to the PBI^{131} value immediately before injection of TSH. In each of the 12 patients studied, blood samples were taken 48 and 24 hours prior to injection of TSH and 0.5, 1, 1.5, 2, 4, 8, 12, 24 and 48 hours after that injection. The broken line represents the mean value for PBI^{131} in 6 patients not treated with TSH.

Release of thyroid hormone however, increased continuously with mounting doses of TSH above 0.025 unit per Kg. (Fig. 5).

DISCUSSION

The observed latent period of eight to ten hours between an intramuscular injection of thyrotropin and its effect on the iodine accumulation in the thyroid is closely consistent with the results of earlier investigations in man (2). A similar latent period has been reported in rats (10, 11), even when the TSH was given intravenously (12). The duration appears to be longer—at least twenty-four hours—in chicks (13–15). In our series, the action of TSH on the release of organic iodine from the thyroid gland, on the other hand, started much earlier and was significant ninety minutes

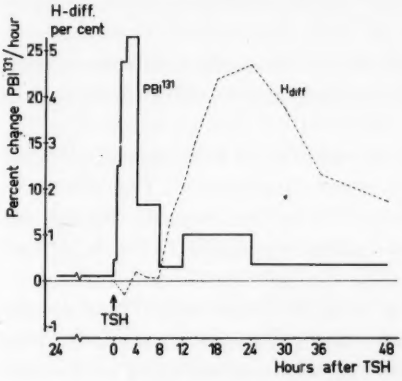
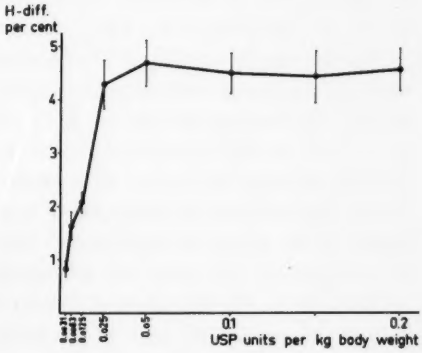
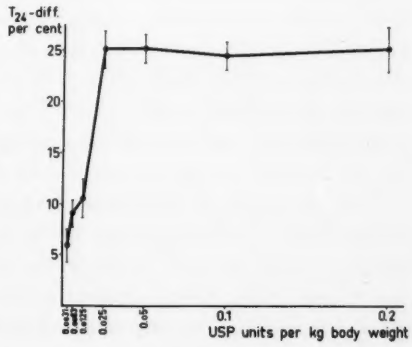


FIG. 3. Time-response relationships after a single intramuscular injection of TSH, showing the percentile increase of PBI^{131} per hour at varying times before and after the injection (*cf.* Fig. 2). The broken line is the time-curve of changes in the thyroidal radioiodine accumulation rate, as measured by H values.



(a)



(b)

FIG. 4. Dose-response relationships after single injections of TSH. **a)** Effect on the thyroidal accumulation of iodine measured by means of the H value, and **b)** by means of the thyroidal 24-hour uptake of I^{131} (T_{24}). The second tracer dose was administered eighteen hours after the injection of TSH. The standard error of the mean is indicated. Twenty patients were tested at each TSH dose level.

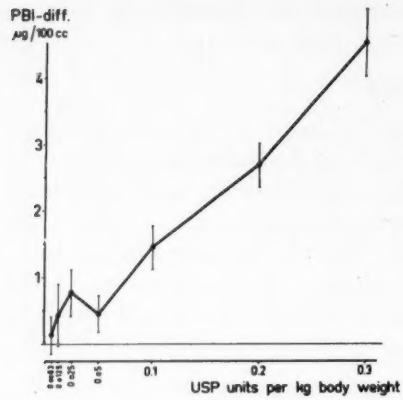


FIG. 5. Dose-response relationships after a single injection of TSH. The effect on release of thyroid hormone was measured by means of changes in serum PBI concentration. Blood samples were taken immediately before the injection of TSH and 42 hours after that injection. The standard error of the mean is indicated. Fifteen patients were tested at each dose level.

after an intramuscular injection of TSH. This earlier effect on release than on uptake has been found in all species studied, and was first described in chicks by Keating *et al.* (13).

The maximum effect of thyrotropin on radioiodine accumulation in the thyroid was observed after eighteen to twenty-four hours. The effect decreased during the second 24-hour period. The fall was already significant ($P < 0.05$) during the first six-hour period of the second day after injection. This held true when the tests were made during thyroid medication, or when only a third of the former dose of TSH was used (7). These results differ from those of previously reported investigations in man. Stanley and Astwood (2), using the accumulation gradient as an index of the rate of radioiodine accumulation in the thyroid, reported that stimulation of iodine accumulation was at its height during the second 24-hour period following injection of TSH. Not until the third day was there a decline towards the pre-injection value.

A rebound inhibition of the iodine-accumulating function of the thyroid was earlier observed by Perlmutter *et al.* (3) in man. The 24-hour thyroidal I^{131} uptake, retested fourteen days after administration of about 24 USP units of TSH, had fallen to levels below the pre-TSH values. The rebound phenomenon is not caused by radiation damage from tracer doses of radioiodine; it occurred when only one dose of I^{132} yielding less than 2 rad in the thyroid was given before the eighth day after injection of TSH (7). The time-curve of inhibition of thyroidal activity after injection of TSH resembles that after a single intravenous injection of thyroxine (16). This lends support to the assumption that the inhibitory effect of TSH on thyroidal iodine accumulation arises from the increased hormone release from the gland after injection of thyrotropin.

That the rate of iodine accumulation in the thyroid can be increased only

to a limited degree by a single injection of TSH is to be expected, and has previously been described in chicks (13, 15) and rats (11, 17–19). Keating *et al.* (10) observed in chicks that the weights of the thyroid glands and the mean acinar cell height even continued to rise with mounting dosages of TSH—above those resulting in maximal stimulation of thyroidal I^{131} uptake. The same was found in rats by Ghosh *et al.* (17). Piotrowski *et al.* (20) observed in chicks that the thyroid weight rose with increasing doses of TSH reaching even above the level resulting in maximal stimulation of iodine release. In the present investigation, thyroid hormone release also increased continuously with mounting doses of TSH to above the level resulting in maximal stimulation of the iodine accumulation rate in the thyroid gland. As regards the thyroidal response to stimulation with increasing doses of TSH, different end-reactions show plateaus at different dose-levels. It seems likely that with increasing doses of TSH the end-reactions approach asymptotic maximal values in this order:

1) rate of iodine accumulation in the gland;
2) release of organic iodine from the gland; and
3) thyroid growth (mean weight or growth of thyroid epithelium).

The thyroidal P^{32} uptake in chicks reached its maximum at the same dose level as did the epithelial response (21, 22); this dosage was higher than that resulting in maximal stimulation of the I^{131} uptake (14).

In view of the large doses of TSH reported in clinical use, it is remarkable that maximal stimulation of thyroidal I^{131} uptake has been obtained after TSH dosages of 0.025 USP unit per Kg. of body weight, corresponding to 1.5 USP units in a patient weighing 60 Kg. However, the results obtained in subjects without thyroid disease should not, without further clinical trials, be applied to patients with thyroidal disorders. The results nevertheless suggest that considerably smaller doses than those hitherto used may be effective clinically (7).

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THE THYROID-STIMULATING HORMONE CONTENT OF THE HUMAN HYPOPHYSIS: A PRELIMINARY REPORT*†

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ABSTRACT

The thyroid-stimulating hormone (TSH) content was measured in 203 pituitary glands—198 obtained at autopsy and 5 obtained at operation. In 6 pools composed of 154 glands (total), the mean TSH content ranged from 46 to 155 U.S.P. milliunits (m.u.) per gland. In 44 individually assayed hypophyses obtained at autopsy, the mean TSH content was 114 ± 15.9 m.u., with a range of 10 to 477 m.u. In 5 individually assayed glands removed at operation, the TSH content ranged from 220 to 1100 m.u. In 11 patients less than 51 years old, the mean TSH content of the pituitary was 210 m.u., whereas in the patients more than 51 years old the mean TSH value was 82 m.u. In 13 patients with jaundice of more than four days' duration, the TSH content of the pituitary was 74 m.u.; in the 15 most cachectic and chronically ill patients it was 55 m.u. (below average); in 7 who died acutely it was 196 m.u. In 2 patients receiving prednisone, the pituitary TSH values were the lowest observed in the series. In 9 patients with thyroid nodules the TSH content of the pituitary was low (38.2 m.u.), and in 4 with thyroid atrophy it was high (172 m.u.). In a single untreated thyrotoxic patient, the TSH content of the pituitary was the highest of any (1100 m.u.), suggesting that in this case the thyrotoxicosis may have resulted from pituitary hyperfunction. There was no apparent correlation between the TSH content of the pituitary and any of the following: body weight, length of time before death and autopsy, the hour of the day when death occurred, adrenal histologic picture, and such conditions as cirrhosis without jaundice, uremia, fever, and altered electrolyte levels.

THE presence of thyroid-stimulating hormone (TSH) in the human pituitary gland was first demonstrated in 1935 by Müller, Eitel and Loeser (1). They assayed 74 glands obtained at autopsy. Using a guinea-pig assay method in a group of 53 acetone-dried glands, they obtained a mean TSH content of 26.9 arbitrary units per gland (range, 5.2–86.0 units); in a second group of 21 glands handled differently, they obtained a

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mean TSH content of 12.1 arbitrary units (range, 5–25 units). None of their patients had pituitary, thyroid or adrenal disorders and the only correlation between hypophyseal TSH content and clinical data was a suggestion that a greater than normal amount of TSH was found in pituitaries of patients succumbing to infectious disease. In 1938 Cope (2) confirmed qualitatively the presence of TSH in a pool of 6 human pituitaries. Subsequently, Kunkel and Loeb (3), Chance, Rowlands and Young (4) and Witschi and Riley (5) reported assays of human material, but presented no quantitative data.

In 1954, Burt and Velardo (6) studied 18 pituitary glands removed at autopsy. Half of each gland was examined histologically and half was assayed for TSH by injection into hypophysectomized rats. Only 3 of these 18 pituitaries unequivocally contained TSH. These 3 patients had thyroid glands that were histologically more active than those obtained from the remaining 15 patients. One of the 3 had had thyrotoxicosis treated with radioiodine and at autopsy had a 75-Gm. multinodular goiter.

Two years later the same laboratory reported (in abstract) the assay of 16 pituitaries (7). Using a more sensitive assay for TSH (thyroidal I^{131} uptake in the day-old chick) they found that in 1 patient with myxedema who was receiving thyroid therapy terminally there was no elevation of pituitary TSH; in 3 patients with high TSH values there was associated evidence of decreased gonadal activity; and in 2 with very low TSH values, the patients had received cortisone therapy. From these few assays they made no correlation with age or sex. A full report of this study is in preparation (8).

In 1956 Currie *et al.* (9) assayed extracts of 12 adult human pituitary glands divided into 3 pools. Using the Heyl-Laqueur method the TSH potencies of the pools were closely similar. This is the first report to estimate TSH values in terms of a standard reference substance.

Only the last of these 9 reports included a TSH standard, and none gave sufficient data to permit estimating the precision of the reported values. Because of this paucity of data we began the present study.

MATERIALS AND METHODS

Of the 208 pituitary glands studied, 198 were obtained at autopsies performed an average of twenty-five hours (range, 1.5–105.0 hours) after death, and 5 were obtained at operation.[1] Forty-four of the glands obtained at autopsy were selected for individual assay; they were from men aged 25 to 86 (average, 60) years. The 5 glands obtained at operation were from women aged 36 to 66 (average, 51) years. The glands were trimmed

[1] We wish to acknowledge the generosity of Dr. David Becker of the New York Hospital for arranging to obtain these glands and transport them in a frozen state to Seattle.

weighed, and homogenized at 4° C. Aliquots of the crude homogenate were frozen and assayed an average of forty-three days (range, 1 to 102 days) later.[2] In preparing pools, groups of glands were homogenized and extracted by the method of Cierezko modified by Heideman (10). This extract was found to contain approximately half of the original TSH activity.

All assays were performed by the method utilizing the weight change of surviving bovine thyroid slices (11). In this assay, inhibitors are encountered which prevent its direct application to serum and urine but no inhibition or alteration of the slope of the assay response has been observed when measuring any pituitary preparation. Each unknown was assayed with a TSH standard, and both were assayed simultaneously at three or four dose levels at threefold intervals with at least 5 separately incubated slices per point. All slices in any one assay were obtained from a single gland. The TSH potency and fiducial limits were calculated by the methods of Bliss (12). Whenever the fiducial limits were unsatisfactory the assay was repeated. The results are expressed in U.S.P. milliunits (m.u.) per gland.[3]

The TSH content of the individual pituitary gland was considered in relation to the clinical diagnosis, the age, weight and body surface area of the patient, hormonal therapy, the duration of the fatal illness, the presence of jaundice of more than four days' duration, anoxia, uremia, fever, electrolytic changes, the hour of death, the delay between death and autopsy, and the size and histologic structure of the adrenal and thyroid. The histology of the thyroid was studied and graded independently by 3 observers without reference to other data. When samples for pituitary histologic sections were taken, a scrupulous attempt was made to divide the gland sagittally or, if it had been removed in pieces (as is usual at operation), to cut each fragment in half, since the concentration of TSH may vary widely in different portions[4] of the gland (13, 14). In the 2 surgically removed glands from which histologic samples were taken before weighing, an estimated weight was used to calculate the TSH content.

RESULTS

In Table 1 are presented the TSH assay data for the 198 glands obtained at autopsy. Only the first 3 pools (a total of 70 glands) were known to be

[2] Repeated assay of a pool of homogenized pituitary tissue for TSH revealed a decline from an initial pre-freezing potency of 10 U.S.P. m.u./mg. (95 per cent fiducial limits, 7.1–14.0) to 7.6 m.u./mg. (5.8–9.9) after being frozen one day; to 7.5 m.u./mg. (4.8–12) after six weeks; to 4.6 m.u./mg. (3.5–5.6) after twelve weeks; and to 2.5 m.u./mg. (1.7–4.4) after seventeen weeks.

No correction for time in storage was made in the present study because the assays of the 44 individual glands demonstrated no correlation between TSH potency and time in storage; the glands frozen 0–15 days had a mean log TSH content of 0.821, those frozen 16–65 days had a mean log TSH content of 0.949, and those frozen 66–102 days had a mean log TSH content of 0.843. (A log potency of zero is equivalent to 10 m.u.)

[3] Values for the concentration of TSH in glands obtained at autopsy are not presented because they showed a progressive chronologic increase with a commensurate decrease in gland weight as we became more skillful in trimming the gland prior to weighing.

[4] Assay of different portions of 3 pituitary glands from cattle revealed that the anterior peripheral portions contained 11, 41 and 6.6 per cent of the TSH in the respective central portions, and the posterior peripheral portions contained 6.6 and 3.1 per cent of the TSH in the central portions. The posterior lobes contained no detectable TSH.

TABLE 1. TSH CONTENT OF 198 PITUITARY GLANDS OBTAINED AT AUTOPSY

Group & Sex	No. of glands		TSH (m.u./gland)
	VA Hosp.	King Co. Hosp.	
Individual (male)	44	0	114±15.9*
Pool 1 (male)	24	0	133 [66.6–310.6]†
Pool 2 (male)	26	0	155 [85.3–282.6]
Pool 3 (male)	20	0	99 [74.2–131.1]
Pool 4 (male & female)	8	4	94
Pool 5 (male & female)	12	35	46 [11.0–73.6]
Pool 6 (male & female)	33	22	51 [24.0–108.8]

* Mean and Standard Error.

† Estimated TSH content and 95% fiducial limits. The TSH contents of the pools were not corrected for loss during extraction (approximately 50%).

from patients of the same sex (male) and age as those providing the 44 individual glands. The weighted mean TSH content of these 3 pools was 131.4 m.u. per gland, closely similar to the average value for the individual pituitaries. The other 3 pools contained a total of 61 glands from patients in a county hospital. This group included women and more elderly and cachectic patients; the weighted mean TSH content was 83.1 m.u. per gland.

Table 2 shows the concentration and total content of TSH in the 5 pituitary glands surgically removed because of metastatic cancer of the breast. Although all the values were higher than the *mean* autopsy values, one which was obtained in a woman with active thyrotoxicosis was markedly elevated above the others. This patient was mildly toxic, had a dif-

TABLE 2. TSH IN 5 PITUITARY GLANDS OBTAINED AT OPERATION

Patient, No. & sex	TSH (m.u./mg.)	Weight of gland (mg.)	TSH (m.u./gland)
1. F	0.57 [0.23–1.2]*	393	220 [89–460]*
2. F	0.79 [0.36–1.8]	303	240 [110–540]
3. F	0.84 [0.58–1.2]	†	300 [200–430]†
4. F	0.64 [0.44–0.93]	309	250 [170–360]
5. F	3.2 [1.9 –4.8]	†	1100 [670–1700]†

* The 95% fiducial limits of assay.

† A gland weight of 350 mg. was estimated, since an unweighed portion was removed for histologic section prior to weighing.

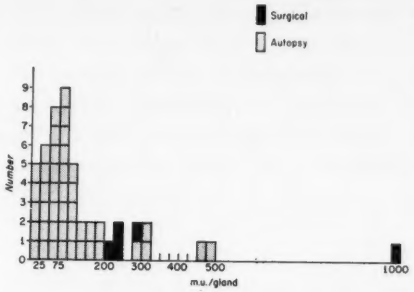

FIG. 1. The TSH content (U.S.P. milliunits) of 49 individually assayed human pituitary glands; 44 were obtained at autopsy and 5 at operation.

fusely enlarged gland but no exophthalmos, and had received no antithyroid therapy for a month prior to surgery.

Figure 1 shows the distribution of values for the TSH content of the 49 individually assayed glands. They ranged from <13 to 1100 m.u. per gland.[5]

Figure 2 shows the average values in certain sub-groups. TSH content appeared to decline with age; in 12 patients less than 51 years old, the average TSH content was 210 ± 45.8 m.u. per gland; in 32 patients more than 51 years old, the average TSH content was 82 ± 10.4 m.u. per gland. In the 13 patients with jaundice of more than four days' duration, the average TSH content was 69 ± 13 m.u. per gland. In 7 persons who died acutely in less than seventeen hours after previous apparent satisfactory health, the average TSH content of the pituitary was 190 ± 40 m.u. The 15 patients with the lowest weight per unit height at the time of death were selected as the most cachectic and chronically ill. Their average weight loss was $41\frac{1}{2}$ pounds during an average illness of 7.8 months. The average TSH content of their pituitary glands was 55 ± 8.1 m.u., a value significantly less than the TSH content in less cachectic subjects. In 2 patients who died while receiving large doses of prednisone for one week and three months, respectively, the values for TSH content of the pituitary were the lowest in the series.

Nine patients had nontoxic thyroid nodules: in 2 the nodules were noted grossly and in 7 they proved to be follicular adenomas upon histologic examination. The average TSH content of the pituitary in these 9 patients was 38 ± 6.8 m.u. Four patients with thyroid atrophy (both grossly and histologically) had high pituitary TSH values (172 ± 49) m.u. although they

[5] A chi-square test for goodness of fit showed ($P < 0.0005$) that the TSH content of the individually assayed 44 glands obtained at autopsy did not follow the normal probability distribution. The same test applied to the logarithms of the potencies showed no significant deviation from the normal probability distribution. Thus, assessment of the significance between groups was calculated from log-potency, not potency data.

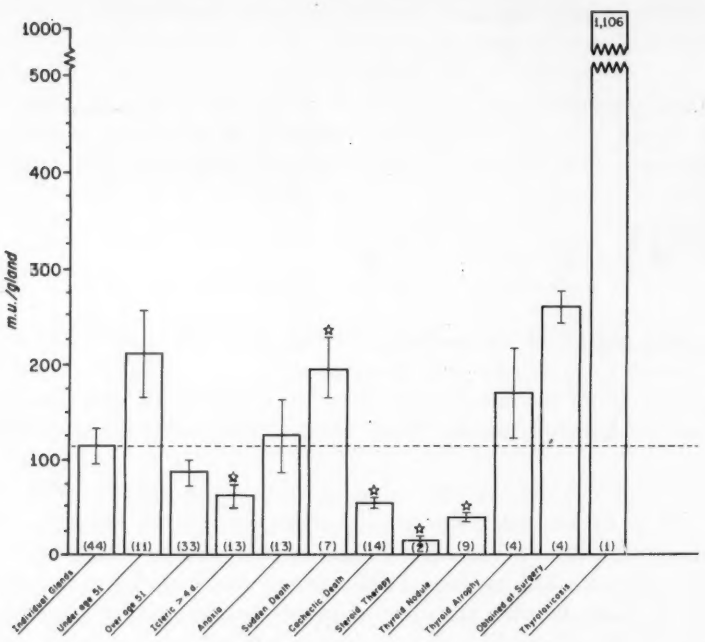

FIG. 2. The TSH content (U.S.P. milliunits; mean and S.E.) of 49 human pituitary glands. The broken line indicates the mean TSH content of the 44 individually assayed glands obtained at autopsy. The number of patients in each subgroup is indicated in parentheses. Significant ($P \leq 0.05$) differences between subgroups and the remainder of the autopsy glands are indicated by (☆). Significance calculated from log potencies (see text, footnote 5).

had not been clinically recognized as having thyroid deficiency. Eliminating these 4 cases, the remaining 40 in the series were divided into categories according to estimates of the height of the epithelial cells of the thyroid. In the 9 cases in which the thyroid cells had flat inactive epithelium, the average TSH content of the pituitary was 51 m.u.; in the 12 with normal cuboidal epithelium the average pituitary TSH value was 109 m.u.; and in the 4 with more active thyroid epithelium the average TSH value was 190 m.u. In the remaining 15 cases, the thyroids were too variable in appearance to be placed in one of these categories.

Because of the small number of patients studied it is advisable to minimize the significance of the average values. However, statistically significant differences ($P < 0.05$) were obtained between the log potencies in certain groups, as indicated in Figure 2. An apparent correlation between TSH content of the pituitary and body weight or body surface was not

statistically significant. There was no apparent correlation between TSH content and any of the following: length of time between death and autopsy, the time of day at which death occurred, histologic structure of the adrenal, and such conditions as cirrhosis without jaundice (3 cases), uremia (10 cases), chronic anoxia (10 cases), fever on the day of death (8 cases), hyponatremia (21 cases), and metastatic malignancy in the thyroid gland (4 cases).

DISCUSSION

The decline in pituitary TSH content with age is consistent with results of animal experiments summarized by Adams (15); there was a significant decline in pituitary TSH content in post-mature aging cattle, rabbits, rats and mice. More recently, Armstrong and Hansel (16) have confirmed this observation in 72 heifers slaughtered according to six advancing age groups over an 80-week period. The declining pituitary TSH content was significantly correlated with declining thyroid weights and epithelial cell heights in these animals. It is of interest to associate these observations with reports that in the elderly there is a decline in the size of the thyroid and a fall in the basal metabolic rate, serum protein-bound iodine level and thyroidal I^{131} uptake (17, 18), and that in senescent rats there is a fall of 20 per cent in the thyroid hormone secretion rate (19). Although the biologic significance of these observations is in doubt (20), the declines may result from a reduction in the secretion of TSH in the aged. Our measurements are compatible either with this hypothesis or with the conjecture that the metabolic need for thyroid hormone is reduced in the aged, so that less need be synthesized and released.

Although the TSH content of the pituitary is not a measure of TSH secretion, it would appear that a decline in pituitary TSH content and thyroid function are parallel in many instances. For example, Adams (15) pointed out that in those animals (*e.g.*, rat, mouse) which normally have very active thyroid glands the pituitary has a high TSH content, and in those animals (*e.g.*, guinea pig, chick) which normally have inactive thyroid glands the pituitary TSH content is very low. The correlation in aging heifers has already been mentioned. In the rat, when thyroid hormone production is blocked, the resulting increase in the release of TSH from the pituitary is associated with a TSH content which rises to persistently high levels lasting many months after a brief initial decline (21). Conversely, when the release of TSH is abruptly suppressed by administration of thyroid hormone, the pituitary TSH content falls to 10 per cent of control values after seven days (22). We have not yet observed any circumstance in which suppression of TSH release was associated with an increase in TSH content, although this possibility remains. Thus, it may be

argued that if thyrotoxicosis were the result of an independently overactive thyroid gland, both pituitary TSH release and content should be low; if thyrotoxicosis were the result of a persistently elevated rate of TSH release from the pituitary, then the TSH content of that gland should be high. The single thyrotoxic patient in this series had an unequivocally high value for pituitary TSH content—suggesting that a high rate of TSH release may have caused her toxic condition. In the 4 patients with thyroid atrophy, who may have had a deficiency of thyroid hormone production, there was also a tendency toward elevated TSH values.

Russfield (8) described hypertrophic hypophyseal amphophils in 6 cases of thyroid failure and in a single case of untreated thyrotoxicosis. Combined with our observations, this leads to the speculation that amphophil hypertrophy may indicate an elevated TSH content and secretion rate in both thyroid failure and in at least some cases of thyrotoxicosis.

A high pituitary TSH content in cases of thyroid atrophy follows the classic feedback theory, but the interpretation of the tendency to low pituitary TSH values in cases of nontoxic nodular goiter is less clear. Possibly a nodule may autonomously produce a normal amount of thyroid hormone, sufficient to suppress pituitary TSH but insufficient to cause toxicity.

The finding of the lowest pituitary TSH values in patients receiving corticosteroid therapy confirms the reports by Russfield *et al.* (7, 8). This observation, considered with reports (23) of a decrease in serum protein-bound iodine concentration, thyroidal radioiodide uptake and thyroid hormone release during corticosteroid therapy, suggests that these steroids may directly depress the pituitary rather than the thyroid. This also agrees with a reported (24) fall in the TSH content of the rat pituitary following administration of ACTH and with the conclusions of Sherer and Siefring (23) regarding corticosteroid therapy in man.

In contrast to the high TSH content of hypophyses obtained after sudden death or during operation, the low TSH values in jaundiced or cachectic patients suggest that in these conditions less TSH is synthesized, possibly as part of a general defect in protein anabolism associated with nitrogen wastage. It is, however, possible that jaundice and its associated liver damage may be more specifically related to the low TSH values. Vannotti (25) reported that in hepatocellular jaundice, thyroxine cannot be inactivated normally because of impaired glucuronide formation. This retention of active thyroid hormone provoked an inhibition of the pituitary gland, manifested by depression of thyroidal I^{131} uptake and a low conversion index. He also stated (26) that a similar sequence occurs late in jaundice due to Laennec's cirrhosis.

In conclusion, it should be emphasized that without determination of

TSH blood levels, measurements of pituitary TSH content alone will not provide the information necessary to settle the issues raised in this discussion. These preliminary results are presented because there is such a paucity of published information about the TSH content of the human hypophysis.

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EFFECT OF DESICCATED THYROID IN LYMPHOCYTIC (HASHIMOTO'S) THYROIDITIS*

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ABSTRACT

A total of 97 patients with nontoxic goiters were treated at the Mayo Clinic with physiologic doses of desiccated thyroid. Of these 97 patients, 36 had histologically proved lymphocytic thyroiditis; 48 were thought to have lymphocytic thyroiditis on the basis of strong clinical and laboratory evidence; 7 had surgically proved adenomatous goiters and 2 others were thought to have this type of goiter; 3 had regenerative hyperplasia after thyroidectomy for Graves' disease; and 1 had proved colloid goiter. Most of the goiters that were caused by lymphocytic thyroiditis responded to treatment with desiccated thyroid, whereas most of those which were adenomatous did not.

DESICCATED thyroid has been advocated for the treatment of goiter from time to time in past years. Greer and Astwood (1) stated that the first published account of administration of thyroid for goiter was by Reinhold (2), in 1894. During the next several years after this initial report, more papers appeared on this subject from Europe and a few other parts of the world, but the use of desiccated thyroid for the treatment of goiter gradually was abandoned for the most part.

Currently, interest has been revived in this form of treatment for goiter. In 1953, Greer and Astwood (1) reported that 76 per cent of a series of 50 patients with both diffuse and nodular goiters treated with desiccated thyroid responded with a decrease in size of the goiter, and 40 per cent experienced complete remission. On the other hand, Crile (3) stated that not more than 5 per cent of several hundred nodular goiters treated at the Cleveland Clinic with 2 to 3 grains of desiccated thyroid daily had disappeared, although he considered that the majority did become smaller. Werner and associates (4) stated that the goiters they had observed to dis-

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† The Mayo Foundation, Rochester, Minnesota, is a part of the Graduate School of the University of Minnesota.

appear during treatment with thyroid probably were associated with chronic thyroiditis and were not true nodular goiter, although they did see a shrinkage of 50 per cent in some nodules. One of the major difficulties in evaluating these results is that histologic proof of the particular type of goiter being treated was not obtained. Thus, one cannot say with certainty what forms of goiters did respond to treatment with thyroid and what forms did not.

Statland and associates (5) and Skillern and co-workers (6) reported that treatment with desiccated thyroid greatly decreased the size of goiters proved histologically to be caused by lymphocytic thyroiditis.

We recently have employed desiccated thyroid in the treatment of selected patients with goiter, and this report is an account of our experience with this treatment. We tried to obtain histologic proof of the diagnosis in as many patients as possible; in the others, we attempted to establish the diagnosis as accurately as possible by clinical and laboratory means.

MATERIAL AND METHODS

A total of 97 patients who had enlarged thyroid glands of various types were observed at the Mayo Clinic during a trial of treatment with desiccated thyroid. For purposes of evaluation, the patients were classified in 4 groups.

Group 1 comprised 36 patients proved to have lymphocytic thyroiditis by either needle biopsy of the thyroid or thyroidectomy. Group 2 included an additional 48 persons who also were considered to have lymphocytic thyroiditis but in whom histologic proof was not obtained. The diagnosis in both groups of patients was suggested on clinical grounds by the typical "rubbery" feel of the gland, by the association of thyroidal enlargement with either myxedema or a low basal metabolic rate, or by the association of a high thyroidal uptake of radioiodine (I^{131}) with normal or decreased levels of serum protein-bound iodine. Six of the 36 patients in group 1 had myxedema; uptake of I^{131} was measured in 5 of these 6 and was normal or increased in each instance, ranging from 25 to 41 per cent at six hours and from 40 to 49 per cent at twenty-four hours. Fourteen of the 48 patients in group 2 had myxedema; uptake of I^{131} was measured in 8 of these 14 patients and was low in 6 (6 to 11 per cent in twenty-four hours), normal in 1 (23 per cent) and increased in only 1 (75 per cent).

Group 3 consisted of 9 patients; 7 ultimately were proved at operation to have adenomatous goiters, whereas the condition in 2 was so diagnosed without histologic evidence. Group 4 was composed of 4 patients, 1 of whom had a surgically proved colloid goiter and 3 of whom had typical regenerative hyperplasia after thyroidectomy for exophthalmic goiter.

Of the 97 patients studied, all except 6 were female. The ages varied from 4 to 73 years; 11 patients were less than 20 years old, 9 were more than 55, and the other 77 were between 20 and 55 years of age. The dosage of thyroid was 1 to 3 grains per day of strong, uncoated, desiccated thyroid.[1] The great majority of patients received $1\frac{1}{2}$ grains per day.

[1] This preparation (Parke, Davis & Co.) is 50 per cent stronger per grain than is USP thyroid.

RESULTS

Group 1. In this group of 36 patients whose goiters were proved histologically to be caused by lymphocytic thyroiditis, desiccated thyroid caused disappearance of the goiter in 11 instances, a significant decrease in the size of the goiter in 22, and little or no change in 3 (Table 1). The mean age of these patients was 38 years, with a range of 12 to 66 years. The mean BMR was −10 per cent, with a range of −24 to +5 per cent. Only 3 of these 36 patients had a BMR above the zero level. Six had clinical myxedema. For the 21 patients who had determinations of serum protein-bound iodine (PBI) concentration, the mean value was 4.7 micrograms per 100 ml., with a range of 1.2 to 11.3 micrograms. The 24-hour thyroidal uptake of I^{131} tracer was measured in 17 patients; the mean uptake was 33 per cent, with a range of 10 to 60 per cent. Some of these data are summarized in Table 2.

In all but 1 of the 33 patients in this group in whom treatment with desiccated thyroid caused a significant reduction in size of the goiter, the decrease was readily apparent within six months of the onset of treatment, and in 12 the effect was noted within two or three months. Little change in the goiter was noted in 1 patient during eighteen months of treatment, but by twenty-four months the size of the goiter had decreased by half.

Group 2. In this group of 48 patients who were considered to have lymphocytic thyroiditis but in whom histologic proof of the diagnosis was not obtained, desiccated thyroid caused the disappearance of the goiter in 28, a significant decrease in size in 16, and little or no change in 4 (Table 1). The mean age of these patients was 36 years, with a range of 4 to 59 years. The mean BMR was −11 per cent, with a range of −31 to +15 per cent.

TABLE 1. EFFECT OF DESICCATED THYROID ON VARIOUS TYPES OF GOITERS

Group	Cause of goiter	No. of cases	Effect on goiter		
			Disappeared	Decreased in size	Unchanged
1	Proved lymphocytic thyroiditis	36	11	22	3
2	Probable lymphocytic thyroiditis	48	28	16	4
3	Adenomas	9	0	3	6
4	Regenerative hyperplasia	3	1	2	0
	Increase in colloid	1	0	0	1

TABLE 2. CLINICAL DATA IN GOITROUS PATIENTS GIVEN DESICCATED THYROID

Group	Cause of goiter	No. of cases	Mean values			
			Age (yrs.)	BMR (%)	PBI (μg. per 100 ml.)	24-hr. I^{131} uptake (%)
1	Proved lymphocytic thyroiditis	36	38	−10	4.7	33
2	Probable lymphocytic thyroiditis	48	36	−11	4.2	26
3	Adenomas	9	43	− 9	4.6	32

Only 4 patients had values for the BMR above zero. Serum PBI concentration was measured in 22 of the patients; it ranged from 0.9 to 6.7 micrograms per 100 ml., with a mean of 4.2 micrograms. Fourteen of these 48 patients had myxedema. The thyroidal 24-hour uptake of a tracer dose of I^{131} was measured in 19 patients; it varied from 6 to 75 per cent, with a mean of 26 per cent. Some of these data are summarized in Table 2.

As in group 1, if treatment with desiccated thyroid caused significant reduction in the size of the goiter, the decrease was readily apparent within six months. The effect in most of these patients was noticeable between two weeks and four months after use of thyroid was begun.

Group 3. Six of the 9 patients with adenomatous goiters who received desiccated thyroid for two to twelve months showed no decrease in the size of the goiter (Table 1). In 2 patients who had large multiple nodular goiters, the treatment brought about a decrease in the over-all size of the goiter but no decrease in the size of the nodules that could be individually palpated. In the ninth patient of this group, treatment with thyroid for three months decreased the size of the goiter by one third. A large single fetal adenoma of the right lobe was found at operation. None of these patients had myxedema.

The ages of the 9 patients varied from 28 to 73 years, and the BMR ranged from −20 to +3 per cent, with a mean of −9 per cent (Table 2). The serum PBI levels, determined in 3 of these patients, were 3.9, 4.5 and 5.4 micrograms per 100 ml.

Group 4. The 1 patient in this group who had a colloid goiter had no significant decrease in the size of the goiter after treatment with desiccated thyroid for nearly three months. This lack of response to administration of thyroidal hormone is surprising. Perhaps regression of the goiter would have occurred if treatment had been continued for a longer period. The

remaining 3 patients in this group had regeneration of thyroidal tissue after subtotal thyroidectomy for exophthalmic goiter but did not become hyperthyroid again; treatment with desiccated thyroid caused disappearance of the recurrent goiter in 1 and pronounced decrease in the size of the goiter in the other 2 (Table 1).

COMMENT

In the great majority of patients who had lymphocytic thyroiditis, treatment with 1 to 2 grains of strong desiccated thyroid caused the goiter to disappear or to decrease in size significantly. Although final confirmation is lacking, this phenomenon is regarded as probably due to inhibition of release of endogenous thyrotropin. The inference that the goiter in lymphocytic thyroiditis is related to increased secretion of endogenous thyrotropin has not yet been confirmed. However, patients who have lymphocytic thyroiditis and normal or increased thyroidal uptakes of I^{131} usually do not show increased uptakes after intramuscular injection of exogenous thyrotropin (6), suggesting that the thyroid already may be under maximal stimulation by endogenous thyrotropin, and hence that such stimulation may be the cause of the goiter.

Recently reported studies (7–10) on the presence of an antibody-antigen reaction in Hashimoto's thyroiditis raise the possibility that treatment with desiccated thyroid might cause a decrease in the size of the goiter by inhibiting the production of antibody and thus decreasing the infiltration of the gland with lymphocytes and plasma cells.

We have been surprised and interested to note the great increase in the number of patients with lymphocytic thyroiditis seen at the clinic each year during the last fifteen to twenty years. Lymphocytic thyroiditis, as defined in this paper, encompasses a wide spectrum of morphologic alterations varying from the classic diffuse infiltration of lymphocytes and plasma cells with associated epithelial involvement to spotty or focal changes of similar type. This concept of lymphocytic thyroiditis has been presented in a current morphologic study of a large group of surgically resected thyroids affected by this disorder (11).

Treatment of patients with adenomatous goiter by administration of desiccated thyroid caused no significant decrease in the size of the goiter in most instances. This is what one would expect if the growth of the usual benign adenoma is not stimulated by thyrotropin but is autonomous. In the 3 patients exhibiting some decrease in the size of an adenomatous goiter while receiving thyroid, the reduction was attributed presumably to lessening in size of the nonadenomatous tissue. The large "adenomas" themselves apparently changed little.

One patient in addition to the 97 patients already considered deserves

special mention at this point. A 34-year-old woman had noted a lump in the left lobe of the thyroid for eight years and a lump in the right lobe for one month. Examination disclosed 2 soft nodules, one 1.5 cm. in diameter in the left lobe and the other 1.0 cm. in diameter in the right lobe. The patient was euthyroid clinically. She was given 1½ grains per day of strong desiccated thyroid, and the gland was re-examined two months later. At this time, the nodule in the left lobe had disappeared but the nodule in the right lobe was unchanged. Shortly thereafter, when thyroidectomy was performed, the left lobe was found to contain the tiny remnant of a benign adenoma; the nodule in the right lobe, which was unchanged by the use of thyroid, contained grade 1 papillary adenocarcinoma.

The difference between the reports of some investigators that almost all goiters respond to treatment with desiccated thyroid and our findings that adenomatous goiters were little affected by such therapy may be due to 2 factors. In the first place, unless tissue is available, one cannot be sure what type of goiter is present. Since histologic identification of the type of goiter being treated was not obtained in previous similar studies, it may be that the majority of such patients had lymphocytic thyroiditis and not adenomatous goiter. In the second place, the correct palpation of the gland is essential for assessment of the result of treatment of a goiter with thyroid. Unless the entire thyroid is carefully palpated from in front of the patient by grasping each lobe separately between the thumb and fingers of one hand, it is difficult to tell whether a gland is nodular or not and whether it has changed significantly in size.

Further confusion may arise from not differentiating between the true encapsulated adenoma, the nodularity of involution following hypertrophy, and the much less common lobulated and nonencapsulated "adenomatosis" of the thyroid. The first 2 types of goiter are little, if any, affected by treatment with thyroid, whereas the last-named form usually is greatly decreased in size by such treatment.

Needle biopsy of the thyroid by means of the Silverman needle has been of great help to us in the confirmation of the diagnosis when the presence of lymphocytic thyroiditis is suspected. It has proved to be a safe and relatively simple procedure, not requiring hospitalization. The thyroid must be sufficiently large to accommodate the needle. Except in extremely small fibrous glands, satisfactory specimens usually may be obtained without difficulty or particular discomfort to the patient (Fig. 1). A single complication, namely hemorrhage into an adenoma, was seen in 1 of approximately 100 patients. After biopsy, this patient complained of increasing pain, which then subsided in three or four days. When thyroidectomy was performed six days after biopsy, moderate extravasation of blood was

FIG. 1. Representative sections obtained by needle biopsy of thyroidal tissue (×2).

found within the capsule of the adenoma from which the specimen had been removed.

The histologic examination of tissue removed from the thyroid by needle biopsy, during which it is preferable to obtain one or more cores of tissue from each lobe, has been found to be reasonably satisfactory (Fig. 2). The limitations of this method as compared to subtotal surgical resection and histologic examination of multiple blocks of tissue are obvious. Needle biopsy in classic diffuse thyroiditis with lymphocytic and plasma-cell infiltration and pronounced epithelial changes should involve no difficulty. Spotty changes of similar type are usually diagnostic upon needle biopsy, although the degree and extent of the process cannot be accurately evaluated. The greatest difficulty in interpretation occurs with the fairly infrequent, extremely fibrotic thyroid with little or no epithelial tissue.

It has not been our purpose to attempt to differentiate malignant from benign nodular goiters by means of needle biopsy but only to confirm the

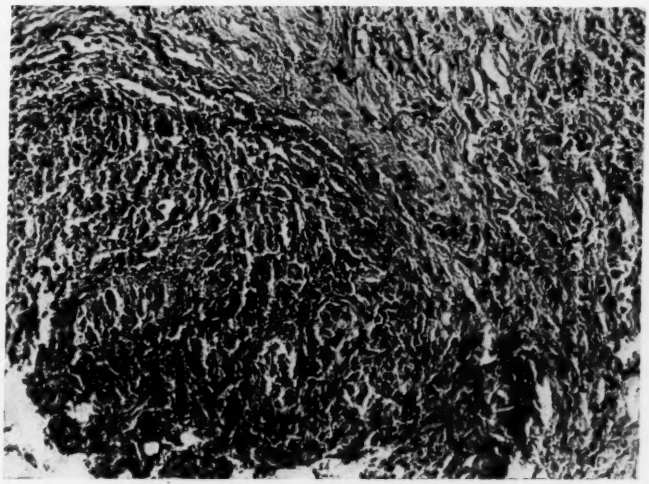

FIG. 2. Tissue removed by needle biopsy in a case of lymphocytic thyroiditis (hematoxylin and eosin; ×150).

diagnosis of lymphocytic thyroiditis. Carcinoma or lymphosarcoma can coexist with lymphocytic thyroiditis, although this combination is rare. Therefore, the physician who elects to treat lymphocytic thyroiditis medically must observe the behavior of the goiter from time to time to make sure that regression is occurring as expected.

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STRUMA LYMPHOMATOSA (HASHIMOTO'S THYROIDITIS) AND RELATED THYROIDAL DISORDERS*

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ABSTRACT

A clinicopathologic study has been made of 605 cases of thyroiditis of the "Hashimoto" type seen during a 27-year period at the Mayo Clinic.

The lesions were divided on a morphologic basis into diffuse thyroiditis (188 cases), thyroiditis with "hyperplastic" epithelium (57 cases) and focal thyroiditis (360 cases). Four histologic subgroups of diffuse thyroiditis were observed, namely, those with extensive epithelial destruction (29 cases), those with oxyphilic epithelium (36 cases), those with varied epithelial changes (106 cases) and those with severe lymphoid infiltration or associated lymphosarcoma (17 cases). These subgroups, as a whole, represent an advanced stage of the disease process. Clinical and laboratory data are provided for these arbitrarily chosen groups, as well as results of thyroidal function studies in a limited number of cases in each category.

These 605 cases included 18 cases of associated carcinoma and 12 cases of associated lymphosarcoma. All the carcinomas, both large and small, were papillary and of low malignancy but showed definite invasion of adjacent parenchyma.

STRUMA lymphomatosa, first recognized in 1912 (1), has shared with other forms of thyroiditis considerable confusion and controversy with regard to definition and nomenclature. In the absence of an etiologic classification, many authors have been inclined to describe and discuss a group of arbitrarily selected cases, particularly those in which diffuse oxyphilia is present, and have tended to neglect the more numerous cases in which lesser degrees of morphologic changes are present, relegating the latter to a rather vaguely described group frequently called "chronic nonspecific thyroiditis." This lack of a clear-cut definition of struma lymphomatosa has created difficulties in assessing its incidence as compared to that of other forms of thyroiditis and in determining its relationship to carcinoma

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or to Graves' disease. It also creates problems in judging whether or not the "disease" is increasing in incidence.

Because of the current interest in many aspects of the problem of struma lymphomatosa and particularly since recent articles (2, 3) still show considerable variation in terminology and definition, a review of our material was undertaken from the clinicopathologic point of view.

MATERIALS AND METHODS

All cases of thyroiditis seen at the Mayo Clinic over a 27-year period (1930 through 1956), in which thyroidal tissue was available, were selected for study. Twenty cases of invasive fibrous thyroiditis (Riedel's struma) and 108 cases of granulomatous thyroiditis (de Quervain's thyroiditis) seen during this period have been reported previously (4, 5).

Approximately 600 cases remained in which "thyroiditis" was the major pathologic diagnosis or in which study of the specimen revealed significant changes of this type, in addition to some other pathologic findings in the gland. Thus, the presence of an adenoma or a carcinoma in one portion of the tissue did not exclude the case from this study, provided the degree of thyroiditis present was moderate to severe. Because of the large bulk of material involved, a special study of cases clinically diagnosed as Graves' disease and in which some degree of "thyroiditis" was mentioned in the original pathologic report was not made, although a few such cases are included, especially those in which goiter recurred after previous thyroidectomy.

All gross specimens of thyroid were available for study, and new blocks were cut from the entire series to provide an adequate sampling of tissue. Cases in which only needle biopsy was performed were excluded; these cases and a large group in which needle biopsy was performed subsequent to 1956 and in which special studies have been carried out will be reported in a subsequent paper. The present study encompasses a total of 605 cases.

PLAN OF STUDY

The plan of study adopted was to classify into subgroups all cases in which the lesions had approximately a similar morphologic picture and to outline the clinical and laboratory data currently available in each. The present study constitutes an attempt at devising a morphologic framework to serve as a basis for current or future data.

Pronounced variation in morphologic changes was seen in this large group concerning the type of "inflammatory" infiltrate, the degree of follicular change, including damage or destruction of follicular epithelium, the amount of fibrosis, and the number of localized lymphoid follicles, or germinal centers. The one most distinctive histologic feature that apparently was common to all the cases in which the condition was advanced was the interfollicular infiltrate composed of plasma cells and lymphocytes. Interfollicular plasma cells were present in all such cases and often appeared to outnumber the lymphocytes. This is of interest in connection with the current theory of auto-immunization and production of antibody within the thyroid.

The arbitrary grouping of cases was based on 1) the degree of the interfollicular infiltration (diffuse or focal), and 2) the changes in the follicular epithelium, including apparent damage or destruction. For the purposes of this study, "diffuse thyroiditis" implies an interfollicular infiltration that was complete, or virtually so, in all zones sampled. In "focal thyroiditis," this change was spotty in character, with larger or smaller islands of parenchyma free of infiltrate. Since, in the past, stress has been placed on the

occurrence of oxyphilic epithelium, a subgroup of cases with "diffuse" lesions in which oxyphilia was uniform and advanced was decided on mainly for comparison with other cases in which equal amounts of diffuse infiltration were present but without the advanced epithelial changes. The lesions designated as "hyperplastic" were chosen because of the uniform epithelial changes; some variation in the degree of infiltration was noted in this group. Because of interest in the problem of primary lymphosarcoma of the thyroid, a small subgroup of cases in which extreme and diffuse lymphocytic or plasma-cell infiltration was present were segregated as the "lymphoid" type, in addition to those cases in which a diagnosis of localized primary lymphosarcoma of the thyroid was made.

The classification just discussed that was used for this study is summarized in Table 1. It is emphasized that this grouping merely attempts to give the spectrum of morphologic change, together with what clinical and laboratory data were available in this large group of cases, and that no arbitrary decision is attempted as to whether all or only some of the cases should be included under the term "struma lymphomatosa." Little information is available as to possible progression of one morphologic type into another more "advanced" stage, since the performance of multiple biopsy is unusual in this disease.

TABLE 1. CLASSIFICATION OF "HASHIMOTO'S THYROIDITIS"

Diffuse thyroiditis
Pronounced epithelial destruction
Oxyphilic epithelium
Varied epithelial changes
Lymphoid type
Thyroiditis with "hyperplastic" epithelium
Focal thyroiditis

DIFFUSE THYROIDITIS WITH EXTENSIVE EPITHELIAL DESTRUCTION (29 CASES)

These 29 cases were placed in this category because epithelial destruction appeared to be the dominant histologic alteration. This was associated with varying degrees of inflammatory infiltration in all cases. Gross examination of the specimen and the description of the thyroid by the surgeon suggested a diffuse fibrotic process in the gland. It appears difficult to categorize exactly the type of thyroiditis present in these advanced lesions, although an atypical variant of granulomatous thyroiditis (de Quervain's thyroiditis) was strongly suggested in 9 instances despite the absence of definite tubercle formation with giant cells. The remainder of the cases apparently represented merely fibrotic or advanced struma lymphomatosa with pronounced loss of epithelium.

The 9 cases in which the histologic findings suggested an atypical form of de Quervain's thyroiditis are included to emphasize the difficulties in histologic diagnosis sometimes encountered. There were 8 women and 1 man, varying in age from 26 to 63 years. Five of these had symptoms of hyper-

thyroidism, with basal metabolic rates varying from +22 to +63 per cent. Two patients, in addition to hyperthyroidism, had some pain and tenderness in the thyroid. The duration of symptoms varied from one to seven months in these 5 patients. The 4 remaining patients had noted pain and tenderness in the gland for one to six months; 2 were euthyroid and 2 were myxedematous when examined at the clinic.

All 9 patients displayed extensive destruction of thyroidal epithelium; this process was recent and acute in some, whereas in others it was in a later stage, with some foci of small regenerative glands such as noted in the healing stage of granulomatous thyroiditis. Studies employing radioiodine (I^{131}) were not made in these 9 patients.

The remaining 20 cases in this group, apparently representing advanced struma lymphomatosa, included 15 women and 5 men. The ages ranged from 36 to 71 years, with an average of 54 years.

TABLE 2. CLINICAL DATA IN DIFFUSE THYROIDITIS WITH EXTENSIVE EPITHELIAL DESTRUCTION

Case	Sex and age (yrs.)	Thyroidal state	BMR (%)	Serum PBI (μg./100 ml.)	Thyroidal I^{131} uptake (% dose in 24 hrs.)	Size of thyroid		Wt. of tissue removed (Gm.)
						Rt.	Lt.	
1	F51	Myx.	−11	2.5	3	4N*	4N	24
2	F36	Myx.	−13	2.6	—	3N	3N	18
3	F66	Myx.	−16	0.7	0	Enl.	Enl.	10
4	F71	Myx.	−16	2.2	63	4N	4N	29

* N = Normal.

Clinical and laboratory data. Histories of previous goiter were obtained in 19 of these 20 patients. The duration was less than one year in 10 patients, one to three years in 7, and five years in 1; the remaining patient had noted a recent (three-month) increase in the size of a long-standing goiter.

No symptoms were associated with the thyroidal enlargement other than a sense of pressure or fullness in the throat in 3 patients, and slight pain and tenderness in 2 patients, noted four months and one year previously. Twelve of the 20 patients were myxedematous when first examined, and the remainder were euthyroid. Studies of thyroidal function were carried out in 4 of the more recent cases (Table 2).

Surgical and pathologic findings. Thyroidal enlargement, as noted by the surgeon, was usually moderate in degree, most glands being estimated as three to four times normal size; exceptionally, they were five to six times normal. Bilateral diffuse thyroiditis was present in all instances (Fig. 1), and all the glands were described as "very firm," "densely hard," "woody" or "fibrotic." Adhesions to surrounding structures were noted in only 2 of the 20 cases, with no indication of the invasive behavior noted in Riedel's

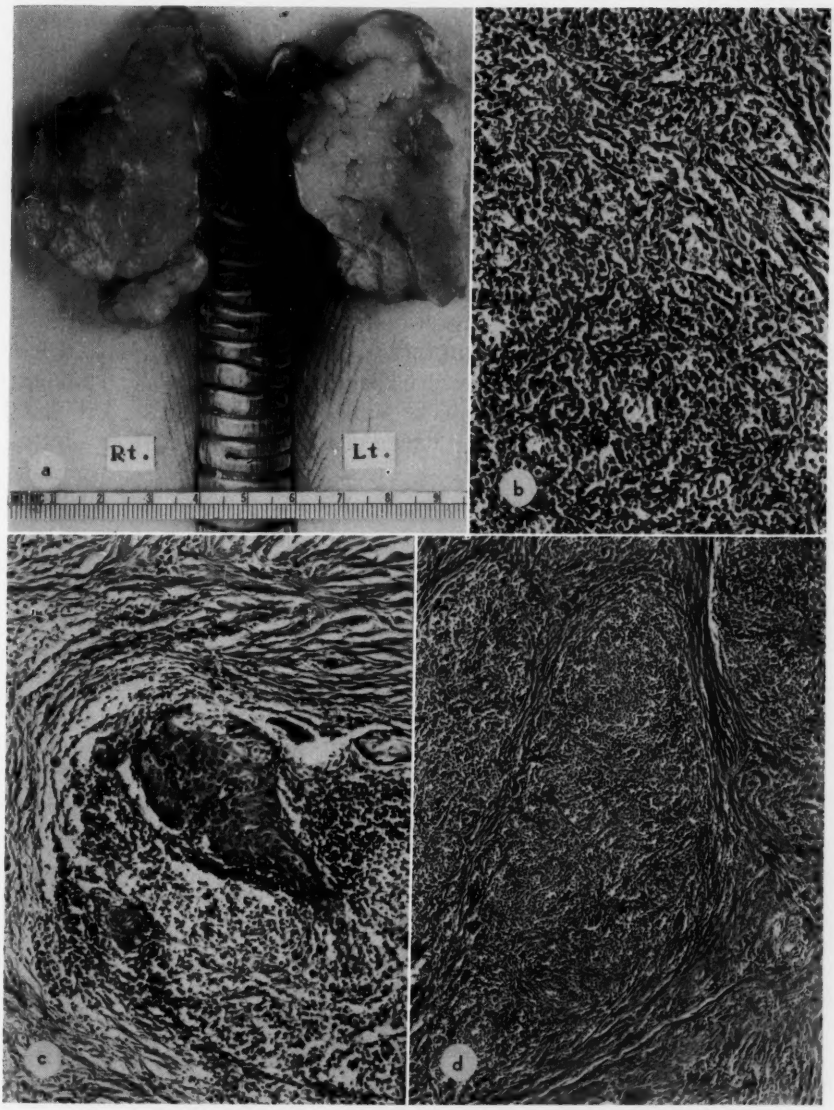

FIG. 1. Diffuse thyroiditis with epithelium largely destroyed. **a.** Gross appearance. **b.** Same case, showing loss of follicles, pronounced inflammatory infiltration and beginning fine fibrosis (reduced from ×170). **c.** Older and more fibrotic type, showing squamous metaplasia of residual epithelium (reduced from ×120). **d.** Lobular pattern outlined by broad bands of fibrous tissue (reduced from ×55). These and all other sections were stained with hematoxylin and eosin.

struma. Bilateral subtotal resection was performed in 15 cases and unilateral subtotal resection in 5.

Histologically, epithelial destruction was the most prominent finding in this group of glands (Fig. 1*b*). Complete destruction was observed in the total sampling in a few cases, but a small number of residual follicles could be found in the majority of instances. In an occasional case, possibly 5 per cent of the total sampled area contained medium-sized foci of residual thyroidal tissue consisting of either small, empty, normal-appearing follicles or small foci of rather hyperplastic or oxyphilic epithelium. Squamous metaplasia of the residual epithelium was noted in half the cases (Fig. 1*c*). Inflammatory infiltration of lymphocytic and plasma cells was noted in all cases. This varied in degree, being pronounced in some lesions and less prominent in fibrotic thyroids in which the process was older. The amount of fibrosis varied somewhat but was prominent in all instances. A distinct lobular pattern outlined by coarse bands of fibrous tissue with fine intralobular fibrosis was a common finding (Fig. 1*d*). This change is comparable to that seen in focal form in a number of cases in the subgroups to be considered later.

Course. Postoperative myxedema was a constant finding in this group, being present in all cases in which follow-up information was available.

DIFFUSE THYROIDITIS WITH OXYPHILIC EPITHELIUM (36 CASES)

These cases were characterized by diffuse interfollicular infiltration with plasma cells and lymphocytes, and diffuse oxyphilic epithelial alteration. The selection was based on a considerable sampling of each specimen (2 to 8 blocks of tissue). Relatively strict histologic criteria were applied, thus excluding cases (see subsequently) in which the oxyphilic change was minimal or focal in character. It is apparent that this "classic" picture is relatively rare.

Age and sex. This group included 33 women and 3 men, a ratio of 11:1. The ages ranged from 35 to 77 years, with an average of 48 years.

Previous goiter. A total of 31 patients gave histories of goiter, with the duration as follows: less than one year, 12 cases; one to two years, 5 cases; three to five years, 9 cases; six to ten years, 3 cases; more than ten years, 2 cases.

Clinical and laboratory findings. In general, most patients had few subjective symptoms referable to the presence of a goiter. Six complained of fullness in the neck, choking sensation or dysphagia. Pain and tenderness were lacking. A total of 27 patients were euthyroid when first examined, and the remaining 9 (25 per cent) were myxedematous. The basal metabolic rates were usually low, being higher than zero in only 4 patients; the

average rate was −8 per cent. Studies of thyroidal function were performed in 8 cases (Table 3).

Surgical and pathologic findings. Bilateral involvement was present in all cases, the degree of enlargement being estimated from two to six times normal, with most thyroids lying midway between these extremes. The capsule was smooth and adhesions were absent. The standard operative procedure was bilateral subtotal resection (29 cases); the weight of resected tissue varied from 3 to 204 Gm., with an average of 64 Gm. Seven patients underwent unilateral subtotal resection, the average weight of the resected tissue being 27 Gm.

Grossly, the thyroids were diffusely enlarged and rather soft, showing no evidence of colloid on cut section (Fig. 2*a*). The lobular pattern usually was

TABLE 3. CLINICAL DATA IN DIFFUSE THYROIDITIS WITH OXYPHILIC EPITHELIUM

Case	Sex and age (yrs.)	Thyroidal state	BMR (%)	Serum PBI (μg./100 ml.)	Thyroidal I^{131} uptake (% dose in 24 hrs.)	Size of thyroid		Wt. of tissue removed (Gm.)
						Rt.	Lt.	
5	M62	Eu.	−12	3.4	25	4N	4N	68
6	F41	Eu.	− 2	3.8	57	2–3N	2–3N	38
7	F59	Eu.	− 5	—	21	4N	4N	99
8	M49	Eu.	0	—	22	2–3N	2–3N	44
9	F37	Myx.	− 8	2.5	31	3–4N	3–4N	87
10	F44	Eu.	− 6	2.4	19	3N	3N	45
11	F35	Eu.	−10	1.8	10	Sl. enl.	Sl. enl.	23
12	F56	Eu.	+ 2	4.0	53 (48 hrs.)	6–8N	6–8N	204

accentuated by thickened interlobular septa. In a few cases (see next paragraph), fibrosis was prominent and the gland was correspondingly firm.

The histologic findings common to all the cases were diffuse interfollicular infiltration and diffuse epithelial oxyphilia (Fig. 2*b* and *c*). The infiltrate consisted of a mixture of plasma cells and lymphocytes in all cases, the former predominating (Fig. 3*a* and *b*). The infiltration was moderate in 10 cases and severe in 26. Localized collections of lymphocytes with germinal centers, although noted in all instances, were not a prominent feature; in only 6 cases were they fairly numerous, being seen occasionally or in widely scattered fashion in the remainder. The degree of fibrosis varied considerably. No increase was noted in 5 cases, and 18 glands showed grade 1 fibrosis, manifested by only slight to moderate fibrous thickening of the interlobular septa. In another 7 cases, fine interfollicular fibrosis was noted in addition to the interlobular fibrosis. The fibrosis was graded 2+ in 5 cases, and it was 3+ in 1 case, with broad septa and considerable focal interfollicular fibrosis (Fig. 3*c* and *d*). Corresponding destruction of epithelium within the fibrous lobules was noted; this was present in foci in 8 cases, 1 of which showed this change in a moderate to severe degree.

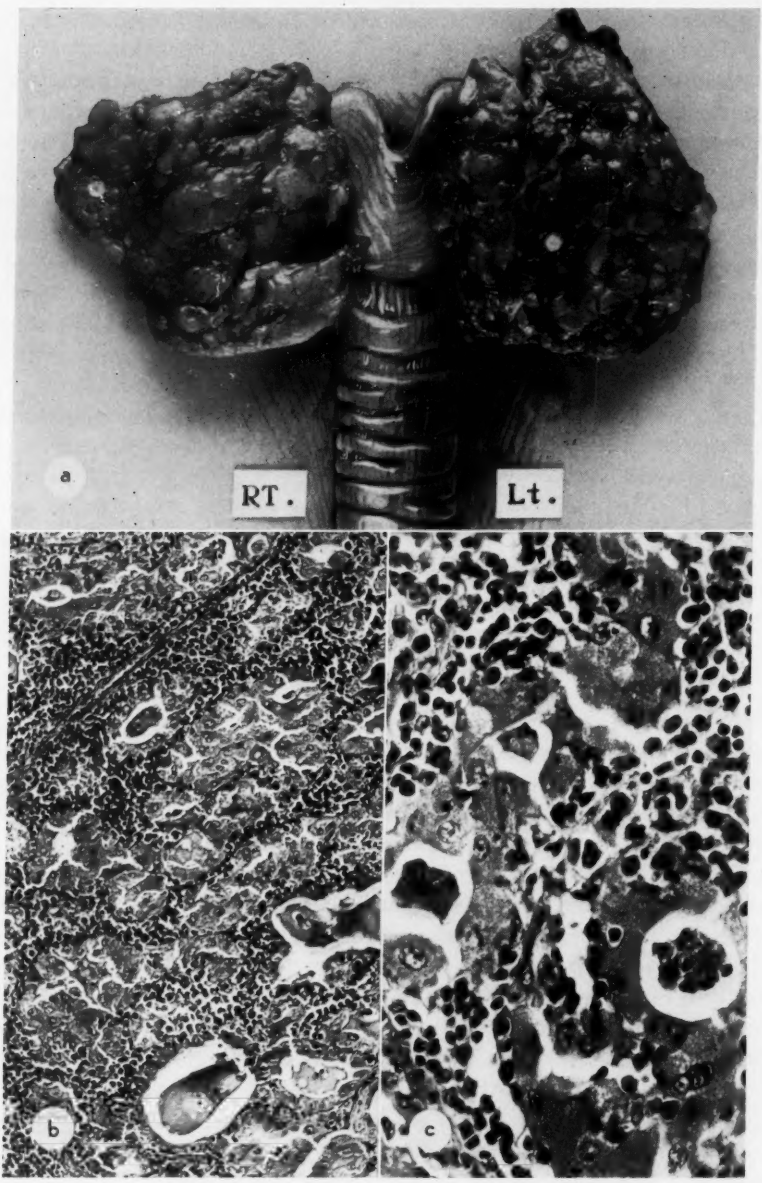

FIG. 2. Diffuse thyroiditis with oxyphilic epithelium. **a.** Note exaggeration of normal lobular pattern. **b.** Diffuse oxyphilia and moderate interfollicular infiltration (reduced from ×150). **c.** Granular oxyphilic cytoplasm (reduced from ×350).

The follicular epithelium was swollen, oxyphilic and granular (Fig. 2*b* and *c*). This change was pronounced in practically all the tissue sampled. The follicles were small to intermediate in size on the average, with the lumens either empty or containing a small amount of inspissated colloid or collections of macrophages (Fig. 3*a*). In many cases, a moderate number of follicles contained a single giant cell in place of the more usual intrafollicular macrophages.

Course. Postoperative myxedema was noted in all cases in which follow-up information was available.

DIFFUSE THYROIDITIS WITH VARIED EPITHELIAL CHANGES (NOT PREDOMINANTLY OXYPHILIC) (106 CASES)

These 106 cases were placed in a separate category mainly for purposes of comparison with those in the oxyphilic group. The lesions, selected histologically, resembled those of the preceding group in having a similar type of diffuse interfollicular plasma cell and lymphocytic infiltration. The epithelium, however, although showing varying degrees of alteration, lacked the diffuse pronounced oxyphilic change previously described. Many of these thyroids did show some oxyphilic epithelium but, when present, it was of rather minimal degree or occurred in a spotty or focal fashion.

Age and sex. This group included 94 women and 12 men, the ratio being 8:1. The ages varied from 19 to 71 years, with an average of 47 years.

Previous goiter. A goiter had been known by the patient to be present before the initial examination at the clinic in 82 of the 106 cases. The duration of the goiter in these 82 cases was less than one year in 25 patients, one to two years in 12, three to five years in 15, six to ten years in 13, and more than ten years in 17.

Clinical and laboratory findings. Symptoms referable to the goiter, when mentioned, usually were minor in character; 20 patients complained of fullness in the neck, a choking sensation or dysphagia, and 7 complained of mild pain and tenderness in the thyroidal region.

On initial examination, 14 patients were thought to have myxedema and 2 were thought to be hyperthyroid; the remaining 90 were euthyroid. The BMR in the 90 euthyroid patients varied from −20 to +17 per cent, with an average of −6 per cent. The average BMR in the 14 patients with myxedema was −16 per cent. Studies of thyroidal function were performed on 18 patients in this subgroup (Table 4).

Surgical and pathologic findings. These were in the main comparable to those in the preceding group. Thyroidal enlargement was usually moderate, the estimate by the surgeon varying from two to eight times normal size. Bilateral subtotal resection was performed in 82 cases; the weight of the re-

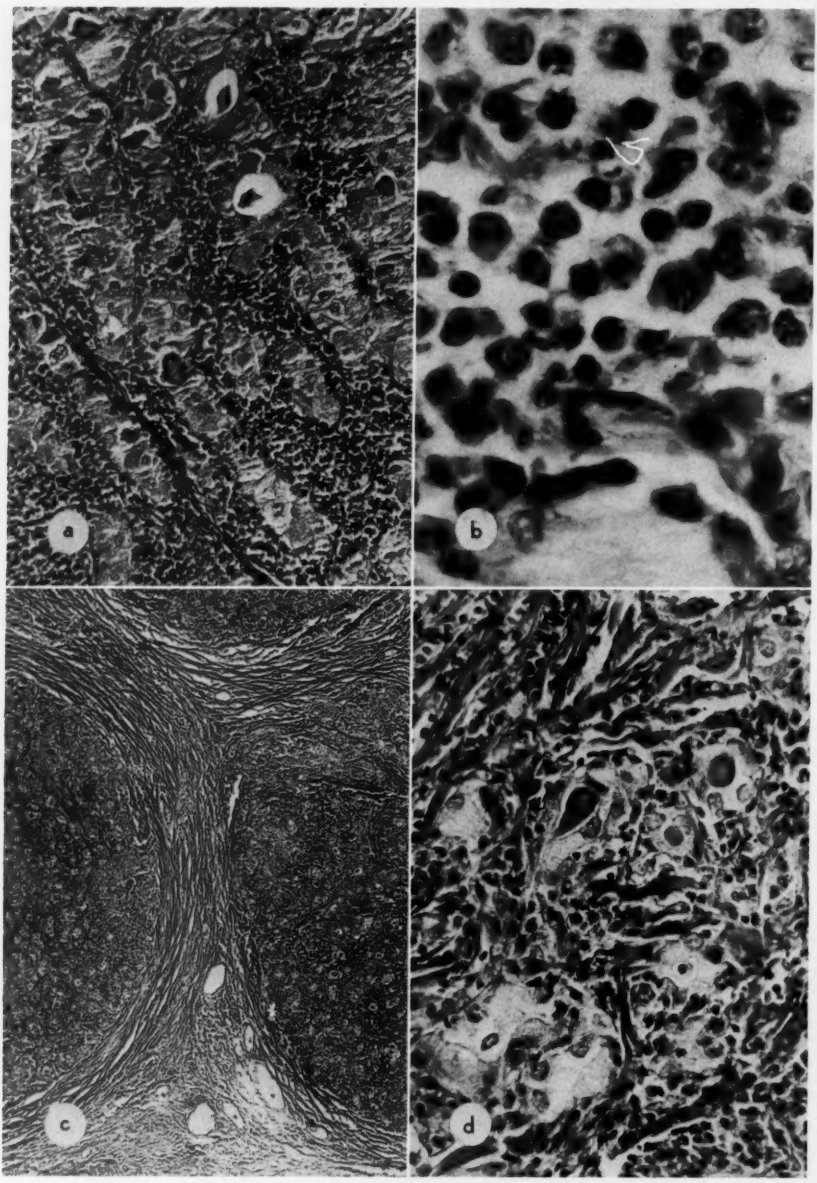

FIG. 3. Diffuse thyroiditis with oxyphilic epithelium. **a.** Severe interfollicular infiltration. Note empty follicles or occasional intrafollicular giant cell (×140). **b.** Same area. Note predominance of plasma cells (×960). **c.** Another case, demonstrating increased fibrosis with broad fibrous septa (×40). **d.** Interfollicular fibrosis in same case. This process results in complete destruction of epithelium in some lobules (×285).

sected tissue varied from 14 to 282 Gm., with an average of 56 Gm. Unilateral subtotal resection was performed in 20 cases, the average weight of resected tissue being 44 Gm. Lesser biopsy procedures were carried out in the remaining 4 cases.

The gross pathologic findings were not appreciably different from those described for the previous group (Fig. 4*a*). Fibrosis was prominent in some.

Considerable variation in histologic findings was observed within this group. As with the oxyphilic lesions, all these thyroids showed more or less

TABLE 4. CLINICAL DATA IN DIFFUSE THYROIDITIS WITH VARIED EPITHELIAL CHANGES

Case	Sex and age (yrs.)	Thyroidal state	BMR (%)	Serum PBI (μg./100 ml.)	Thyroidal I^{131} uptake (% dose in 24 hrs.)	Size of thyroid		Wt. tissue removed (Gm.)
						Rt.	Lt.	
13	F44	Myx.	−10	1.9	51	?	5–6N	40
14	F51	Eu.	− 2	—	42	6–8N	6–8N	150
15	F34	Eu.	− 7	—	34	Recent enl.	Prev. remov.	Not stated
16	F59	Eu.	+16	5.2	49	?	?	16
17	F47	Eu.	−12	—	40	?	?	28
18	F19	Eu.	− 5	6.8	73	3N	3N	29
19	F44	Myx.	− 4	3.6	13	4–5N	3N	88
20	F38	Eu.	+ 2	3.8	19	2½N	2½N	14
21	M49	Myx.	−13	—	57	6N	6N	58
22	F46	Eu.	+ 7	3.8	23	4N	6N	28
23	F57	Eu.	+ 6	2.4	—	Very large	Very large	282
24	F67	Eu.	− 3	2.8	—	Large	Large	59
25	M55	Myx.	−21	2.7	—	2½N	2½N	21
26	F38	Eu.	−25	3.8	20	N	2N	Not stated
27	F51	Eu.	− 5	4.2	—	3N	3N	30
28	F44	Eu.	−20	6.1	52	Large	Large	54
29	F56	Eu.	− 5	4.4	30	2½N	2½N	28
30	F53	Myx.	−25	3.7	21	?	?	25
Range	19–67	5 Myx. 13 Eu.	−25 to +16	1.9 to 6.8	13–73			14–282
Avge.	47		− 7	3.9	37			59

diffuse interfollicular infiltration, with plasma cells a prominent feature in all. This infiltration was, on the average, somewhat less pronounced than it was in the preceding group. It was considered severe in 60 cases (Fig. 4*b*) and moderate in 41; slight, but fairly diffuse infiltration was noted in the remaining 5 cases (Fig. 4*c*). A few glands were included on the basis of a predominantly diffuse infiltrate, although scattered small zones lacked this finding. Localized lymphoid collections with germinal centers were noted in all instances but, as in the preceding group, were not prominent except for 8 cases in which this finding was considered pronounced. The degree of fibrosis was slightly more advanced. No increase was noted in 15 per cent of the cases. Thickening of the interlobular septa was present in 36 per cent, and a degree of associated fine intrafollicular fibrosis as well in another 23

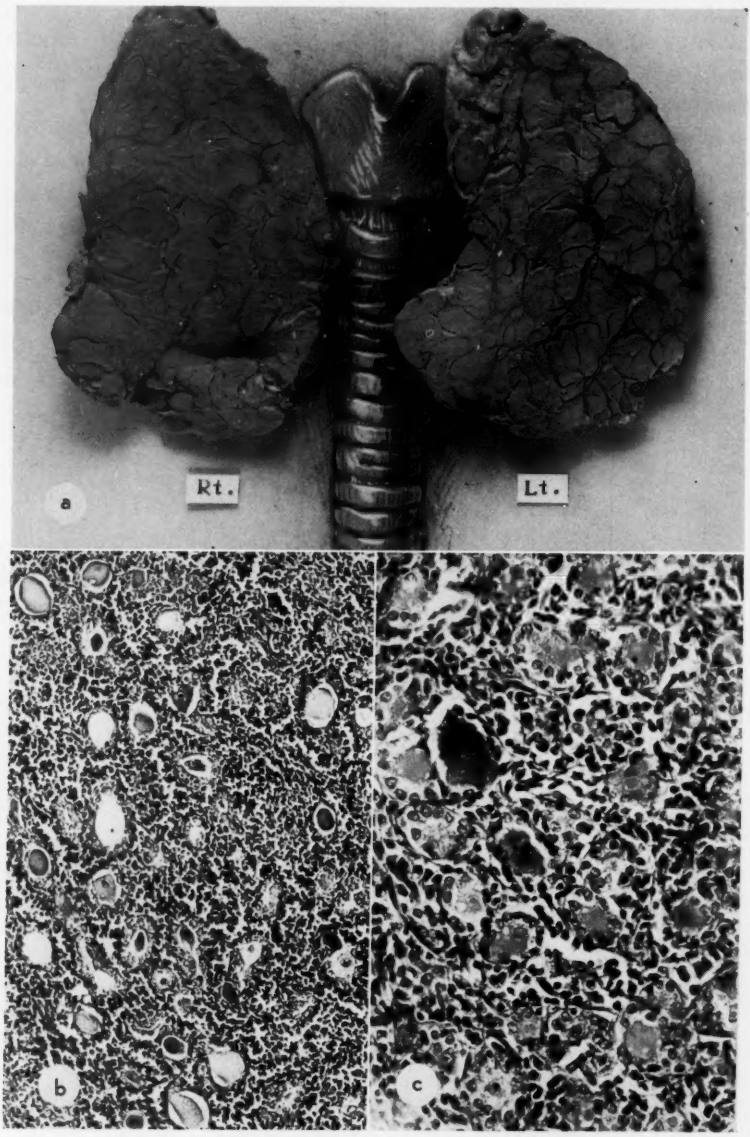

FIG. 4. Diffuse thyroiditis with varied epithelium. **a.** Note distinct lobular pattern. **b.** Same case, showing pronounced interfollicular infiltration and small follicles (×130). **c.** Another similar case, showing slight to moderate interfollicular infiltration; note small cuboidal epithelium (×235).

per cent. Grade 2 fibrosis was noted in 22 per cent of cases and grade 3 in the remaining 4 per cent.

Spotty epithelial destruction occurred in association with the fibrotic process in 16 cases; this was fairly severe in degree in 4 cases. The follicular epithelium and contents showed considerable variation in this group. In general, the follicles were small. A common pattern showed such follicles empty or containing a small amount of inspissated colloid, with occasional small collections of macrophages within the lumens. A mixture of small or intermediate-sized follicles often was noted, with more prominent formation of colloid in the latter. In approximately half the cases, the thyroid also showed foci of oxyphilic cells and, less commonly, focal zones of "hyperplastic" follicles, with more numerous collections of macrophages within the lumens. In a few cases, a more generalized but still focal transition toward the oxyphilic type of lesion was apparent.

Course. Myxedema occurred after bilateral subtotal resection in 98 per cent (58 of 59) of those patients on whom information was available. Among the 24 patients on whom unilateral subtotal resection or biopsy was performed, myxedema developed in 90 per cent (18 of 20) of those followed.

DIFFUSE THYROIDITIS WITH SEVERE LYMPHOID INFILTRATION OR LYMPHOSARCOMA (17 CASES)

Lymphoid infiltration. A surprisingly small number of cases were noted in which the degree of interfollicular infiltration with plasma cells or lymphocytes was of such an unusual degree as to mimic or at least bring up the differential problem of diffuse lymphosarcoma. Only 5 such cases were observed (Table 5). It appears that such lesions are especially prone to occur in elderly women. The gross surgical findings were characteristically those of a medium-sized or large goiter involving both lobes of the thyroid. Bilateral subtotal resection was performed in 4 cases; in the fifth, the operation was left subtotal resection, the right lobe having been removed elsewhere ten years previously.

Gross examination revealed diffuse bilateral enlargement of the thyroid, sometimes unequal in size (Fig. 5*a*). The gland was soft, resembling lymphoid tissue on cut surface, with little or no colloid visible.

Histologic study revealed extensive replacement of thyroidal parenchyma by sheets of lymphocytes in 4 cases (Fig. 5*b*) and by plasma cells in 1 case (Fig. 5*c*). Lymphoid follicles were prominent in this infiltrate in 1 case only and were rare or spotty in the other 4. The cells appeared to be fairly mature plasma cells in 1 instance and mature lymphocytes or lymphoblasts in the remainder. Mitotic figures were readily found (1 or 2 per high-power microscopic field). The amount of residual thyroidal follicular tissue

TABLE 5. CLINICAL DATA IN DIFFUSE THYROIDITIS WITH SEVERE LYMPHOID INFILTRATION

Case	Sex and age (yrs.)	Duration goiter	Symptoms of goiter	Thyroidal state	BMR (%)	Size of thyroid		Wt. of tissue removed (Gm.)	Operation*	Course
						Rt.	Lt.			
31	F61	5 yrs.	Tightness in neck	Eu.	+ 2	3–4N	10N	168	SRLI	Postop. myxedema; well 4 yrs. postop.
32	F49	Many years, with recent increase in size	—	Eu.	+12	5N	3N	53	SRLI	Postop. myxedema; diabetes mellitus developed but otherwise well 9 yrs. postop.
33	F77	1½ yrs.	Pressure in neck	Eu.	−21	4N	4N	112	SRLI	Aplastic anemia requiring transfusions for 2½ yrs. postop.; "malignancy of pelvis" developed 3 yrs. postop.
34	F69	Recent	—	Myx.	−23	—	4–5N	35	SL	Postop. myxedema; well 3 yrs. postop.
35	F63	2 mos.	Fullness in neck	Eu.	—	3–4N	3–4N	30	SRL	Given thyroid postop.; no further data

* SRLI = bilateral subtotal resection with removal of isthmus; SL = left subtotal resection; SRL = bilateral subtotal resection.

varied to a degree, but the tissue was extensively replaced in all instances The appearance suggested gradual destruction or crowding out of follicles by the proliferating lymphoid tissue until, in the most advanced lesions, extremely little epithelium remained in the gland, perhaps 1 or 2 follicles per low-power field in a sea of lymphocytes. However, the process appeared to be incomplete in 2 cases, in which small foci of uninvolved rather normal or slightly oxyphilic thyroidal follicles could be found at the periphery of the gland. It is of interest to note in Case 34 (Table 5), in which recurrence took place ten years after the original lobectomy elsewhere, that the sections from both specimens were practically identical, both showing extreme lymphoid replacement.

The follow-up data in these 5 cases are summarized in Table 5. Insufficient time has elapsed to allow a final decision in all cases, but it appears likely that the process is benign. In this connection, it might be well to record the findings in a case seen at the clinic in 1917 and not included in this series. A 55-year-old woman had a huge bilateral goiter of nine years' dura-

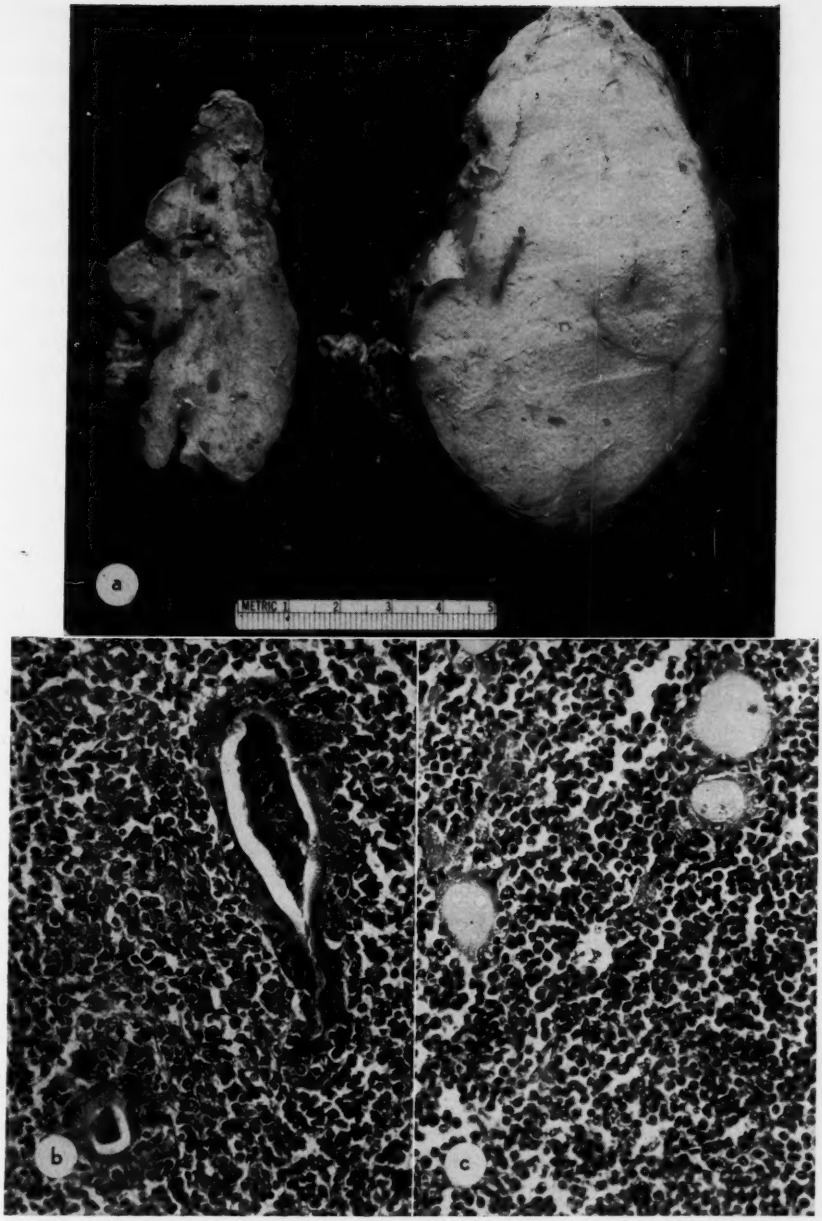

FIG. 5. Diffuse thyroiditis with severe lymphoid or plasma-cell infiltration. **a.** Note absence of fibrosis and the small residual colloid islands. **b.** Same case, showing extensive lymphoid infiltration (×250). **c.** Another case, showing severe plasma-cell infiltration (×275).

tion, which had shown a recent rapid increase in size. Subtotal resection was performed, with removal of 378 Gm. of tissue. The histologic picture showed pronounced plasma-cell infiltration similar to that seen in the cases just described. Irradiation was not given. The patient was alive and well at the age of 83 when last heard from, twenty-eight years after resection.

Lymphosarcoma. In contrast to the 5 cases in which both lobes were diffusely involved and in which at least some follicles remained in the midst of the infiltrate, 12 cases were seen in which a localized lymphosarcoma involved one lobe of the thyroid and in which thyroidal parenchyma was available for study. These cases have been reported previously, along with other cases seen prior to 1930 as well as cases in which biopsy only was performed (6).

The localized tumor, composed of lymphocytes of varying degrees of immaturity, measured from 2 to 10 cm. in diameter and was confined to one lobe of the thyroid. Tissue from the opposite lobe was available in 7 cases and parenchyma from the periphery adjacent to the tumor in the remaining 5 cases; 11 showed a spotty lymphoid change consisting of single germinal centers or groups of germinal centers varying from slight to pronounced in degree. The intervening thyroidal parenchyma was normal or slightly oxyphilic or showed some papillary infoldings. In 3 instances, fairly diffuse interfollicular infiltration with lymphocytes and plasma cells was noted, with a considerable degree of associated oxyphilia. Follow-up data in these cases are provided in the aforementioned paper (6). In general, the outlook is good, especially for those patients in whom extension beyond the thyroidal capsule has not occurred.

THYROIDITIS WITH "HYPERPLASTIC" EPITHELIUM (57 CASES)

It was noted early in the study that thyroiditis in children and young adults frequently presented a rather striking and characteristic histologic pattern that, in its fully developed form, varied little from patient to patient. For this study, all such patients were grouped together regardless of age. In general, the characteristic finding was a "hyperplastic" follicular component showing cuboidal or even slightly columnar epithelium with some irregularity in shape of the follicle or a degree of papillary infolding. Colloid was virtually absent, and intrafollicular macrophages were present in the vast majority of follicles. The degree of interfollicular infiltration with lymphocytes and plasma cells varied from slight to severe in these cases and was not a determining factor in inclusion within this group.

Age and sex. This group included 56 females and 1 male, providing an overwhelming preponderance of females. The ages ranged from 8 to 65 years, with an average of 31 years. The distribution by decades is shown in Table 6.

TABLE 6. AGE DISTRIBUTION IN THYROIDITIS WITH "HYPERPLASTIC" EPITHELIUM

Age (yrs.)	No. of patients
0– 9	2
10–19	6
20–29	19
30–39	18
40–49	6
50–59	5
60–69	1
Total	*57*

Duration of goiter. Histories of previous goiter were obtained in 50 cases. The duration was less than one year in 20 cases, one to two years in 11, three to five years in 6, six to ten years in 7, and more than ten years in 6.

Clinical and laboratory data. Few symptoms associated with the presence of a goiter were noted. Slight pain and tenderness were present in 2 patients and a sense of pressure or dysphagia in 2 others. Six patients were frankly myxedematous or were suspected of having myxedema; the remainder were euthyroid. The BMR varied from −25 to +12 per cent, with an average of −10 per cent. Only 5 patients had a BMR higher than zero.

Additional studies of thyroidal function were performed on 14 patients (Table 7).

TABLE 7. CLINICAL DATA IN THYROIDITIS WITH "HYPERPLASTIC" EPITHELIUM

Case	Sex and age (yrs.)	Thyroidal state	BMR (%)	Serum PBI (μg./100 ml.)	Thyroidal I^{131} uptake (% dose in 24 hrs.)	Size of thyroid		Wt. of tissue removed (Gm.)
						Rt.	Lt.	
36	F23	Eu.	− 4	—	51	5N	5N	32
37	F28	Eu.	−19	3.5	40	?	?	19
38	F22	Eu.	−18	—	35	?	?	29
39	F35	Eu.	−16	—	40 (17 hrs.)	2–3N	2–3N	30
40	F 9	Eu.	−11	5.6	76	4N	4N	27
41	F34	Eu.	− 9	7.3	28	Enl.	Enl.	38
42	F16	Eu.	−14	7.2	—	3–4N	3–4N	30
43	F 8	Eu.	—	4.2	18	2½N	2½N	7
44	F23	Eu.	− 9	—	34	3N	N	19 (rt.)
45	F26	Eu.	− 2	2.2	11	4N	—	40 (rt.)
46	F27	Eu.	− 8	2.9	39	2N	2N	16
47	F20	Eu.	− 2	—	15	5–6N	5–6N	66
48	F23	Eu.	−23	—	41	2N	2N	10
49	F 8	Eu.	− 7	3.1	59	Enl.	Enl.	50
Range	8–35		−23 to −2	2.2 to 7.3	11–76			7–66
Avge.	22		−11	4.5	37			30

Surgical and pathologic findings. The degree of thyroidal enlargement as estimated by the surgeon generally was considerably less than that seen in previous groups, the majority of glands being one and a half to three times normal size. Bilateral subtotal resection was carried out in 45 cases; the average weight of resected tissue was 28 Gm. Unilateral subtotal resection was the procedure in 12 cases, an average of 12 Gm. of tissue being removed.

Gross examination revealed rather small, bilaterally enlarged thyroids without prominent fibrosis and usually devoid of colloid.

The histologic findings were fairly constant in this group, particularly with reference to the follicular changes. Varying degrees of interfollicular infiltration with lymphocytes and plasma cells were noted in all instances. This was slight, and either focal or diffuse in 20 cases (Fig. 6*a*); in the remaining 37 cases, infiltration was either spotty and moderate (18 cases) or diffuse and moderate or severe (19 cases) (Fig. 6*b* and *c*). A rather striking feature was the more prominent formation of lymph follicles as compared to that in the groups discussed previously. In 27 cases, this was extreme, almost every low-power field containing 2 to 4 germinal centers. In 22 cases, these lymphoid collections were considered as moderate, with many low-power fields containing 2 to 3 germinal centers. Lymph-follicle formation was slight in the remaining 8 cases, only an occasional lymphoid nodule being present.

Fibrosis was not prominent. No increase in fibrosis was noted in 20 cases, and slight thickening of interlobular septa was present in 22. In 14 cases, this finding was accentuated by a degree of fine intrafollicular fibrosis that in 1 additional case was graded 2. Focal destruction of epithelium was minimal, being seen in a minor degree in 4 cases.

Associated with the findings just described were follicular changes that were surprisingly uniform throughout the tissue in most cases. As already indicated, most follicles were lined by cuboidal epithelium with some papillary infolding or at least irregularity of follicular outline (Fig. 6*a*, *b* and *c*). The change in 6 cases was minimal, the cells being low cuboidal, whereas columnar epithelium was present in 2 cases. Oxyphilic epithelium was totally absent in most cases; extremely slight focal oxyphilia was noted in 9 cases, and a definite trend toward diffuse oxyphilia was present in only 3 cases. Most follicles contained a small to medium-sized clump of macrophages within their lumens, and fusion of these to form a single giant cell occurred fairly frequently. Some follicles appeared empty, and an extremely small number contained thin colloid. This lack of colloid was a constant and striking feature. In 7 cases in which the morphologic alteration apparently was not fully completed, a few foci of colloid-containing tissue were found. The nature and significance of these intrafollicular

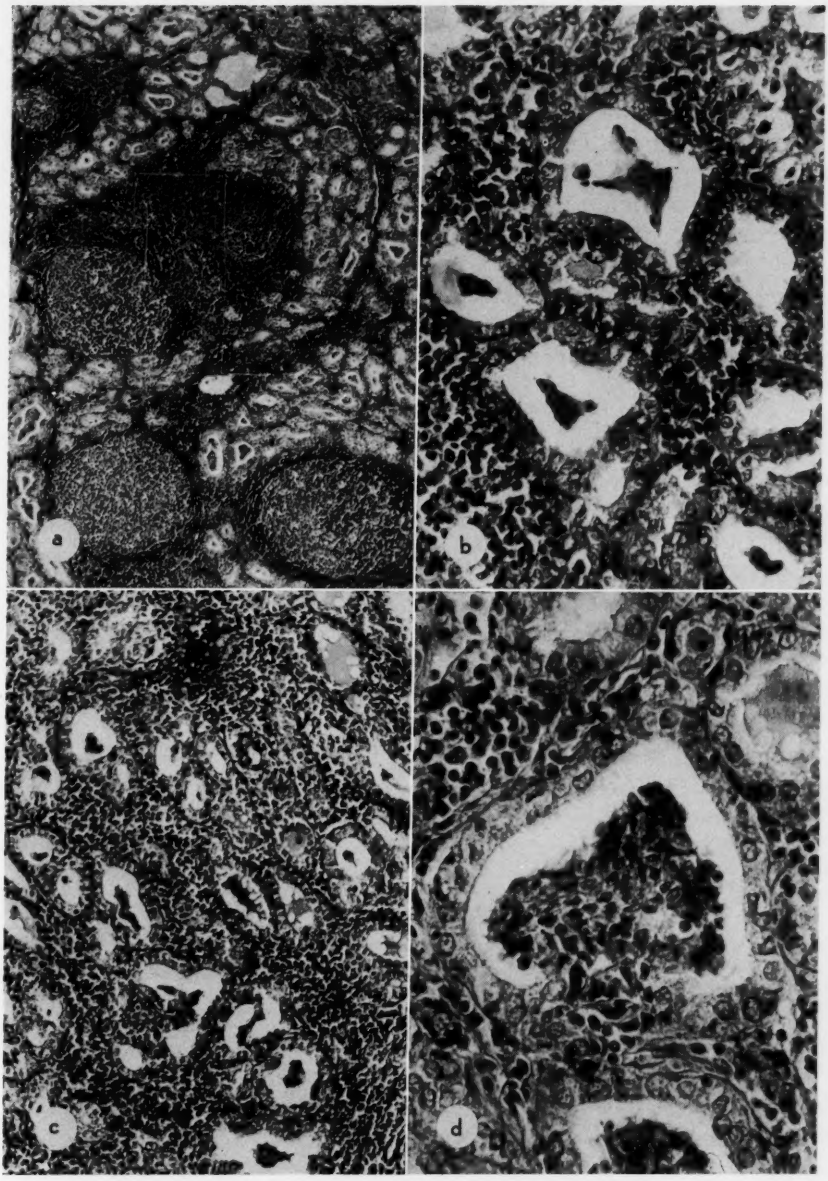

FIG. 6. Thyroiditis with "hyperplastic" epithelium. **a.** Note prominent lymph follicles. **b** and **c.** Similar follicular changes in another case but with moderate and severe interfollicular infiltration (×300 and ×135). **d.** Intrafollicular macrophages. Note penetration of epithelial wall (×390).

macrophages remain a puzzle, although they appear to have some relationship to the functional state of the thyroid. Their point of entry occasionally can be demonstrated by an actual break or defect in the follicular wall (Fig. 6*d*). One patient in this group, a 17-year-old girl, had been treated with 4 grains of thyroid daily for several years without much effect on the goiter. Histologic examination of the thyroid in this case revealed much lymphoid infiltration, with prominent follicles; the epithelium was low cuboidal, and intrafollicular macrophages were scanty or absent in most fields (Fig. 7*a* and *b*).

Course. Follow-up data were available on 41 of the 57 patients. Myxedema developed in 97 per cent of those who had undergone subtotal bilateral resection (33 of 34 patients) and in 4 of 7 patients who had undergone unilateral resection.

FOCAL THYROIDITIS (360 CASES)

A total of 360 patients showed thyroiditis of the type under consideration to a slight, moderate or pronounced degree in which the infiltration was spotty or focal in character. Considerable variation was observed in this group in the degree of infiltration, the type of epithelium and the amount of colloid present in the thyroid.

Age and sex. This group included 343 females and 17 males, the ratio being 20:1. The ages ranged from 10 to 75 years, with an average of 47 years. The age distribution by decades is shown in Table 8.

Duration of goiter. A total of 229 patients had histories of goiter prior to examination at the clinic. The goiter had been present less than one year in 63 cases, one to two years in 36, three to five years in 45, six to ten years in 30, and more than ten years in 55.

Clinical and laboratory findings. Few symptoms were referable to the

TABLE 8. AGE DISTRIBUTION IN FOCAL THYROIDITIS

Age (yrs.)	No. of patients
0– 9	0
10–19	5
20–29	19
30–39	60
40–49	104
50–59	120
60–69	46
70–79	6
Total	*360*

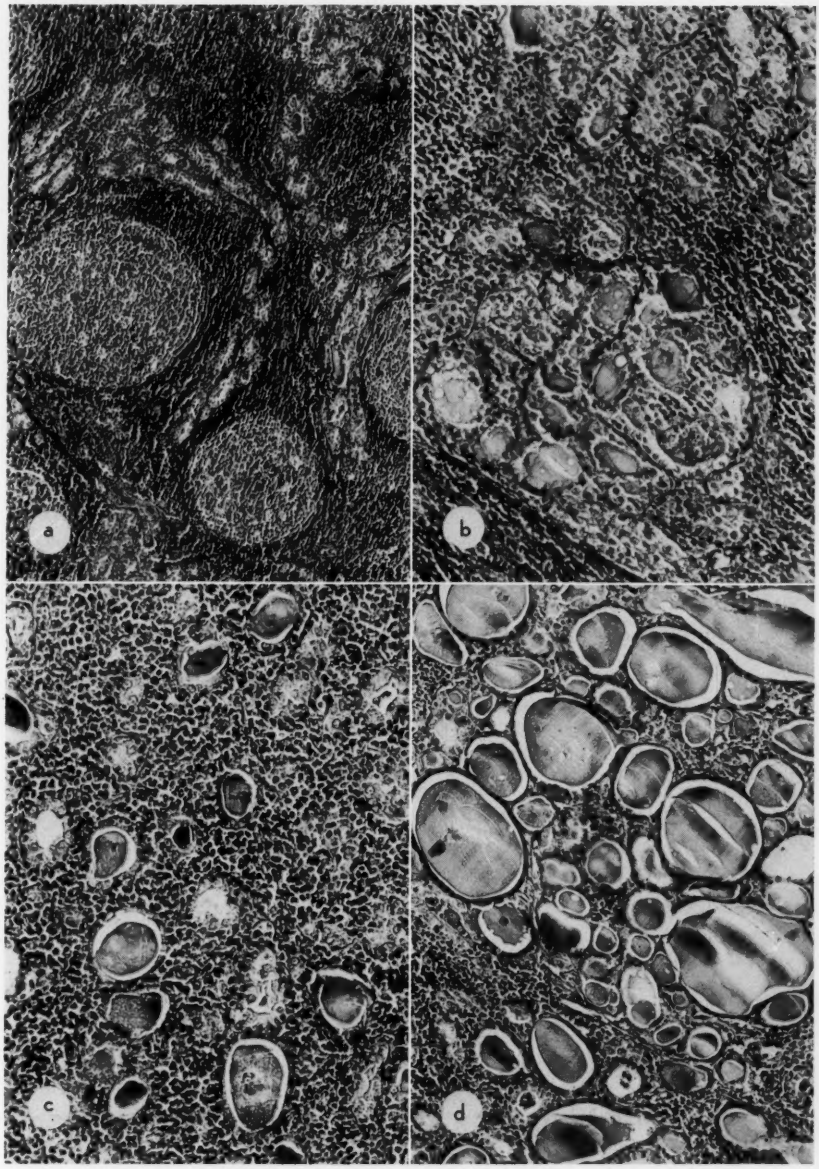

FIG. 7. **a** and **b.** Thyroiditis with "hyperplastic" epithelium treated with desiccated thyroid. There are few intrafollicular macrophages (×65 and ×145). **c** and **d.** Focal thyroiditis. Different regions of same thyroid showing (**c**) severe infiltration in much of gland, and (**d**) one of a number of islands of uninvolved parenchyma (both, ×100).

presence of a goiter, only 25 patients having some degree of pressure sensation or fullness in the neck with occasional dysphagia. Six patients had noted pain or tenderness, usually mild in degree. A total of 344 patients were euthyroid when first examined at the clinic, whereas 9 were hyperthyroid and 7 had myxedema. The average BMR of the euthyroid patients was −4 per cent, with a variation from +18 to −26 per cent. The average BMR of the patients with myxedema was −16 per cent.

Studies of thyroidal function were performed on 29 patients, who had an average BMR of −6 per cent, with a range of −23 to +18 per cent. The serum protein-bound iodine level as determined in 18 patients averaged 4.8 micrograms per 100 ml., with a range of 1.3 to 7.5 micrograms. In only 4 instances was the value less than 3.5 micrograms, the lower limit of normal for our laboratory. Studies of thyroidal iodine uptake were performed on 18 patients; the average uptake was 28 per cent in twenty-four hours, with a range of 7 to 48 per cent.

Surgical and pathologic findings. The gross changes in the thyroid in this group depended on the extent of the disease and, in some cases, evidently on the type of goiter onto which the thyroiditis was engrafted. The size of the thyroid as estimated by the surgeon varied from nearly normal to ten times normal. The greatest weight of resected tissue was 200 Gm., and the average for all cases was 35 Gm. In 210, or almost 60 per cent of the cases, the weight of tissue resected was 30 Gm. or less.

A wide spectrum of histologic changes was noted. The amount of thyroiditis as judged by gross examination and study of multiple blocks of tissue was considered pronounced in 80 cases. In a large number of these, the process was "spotty" in small zones only, the remainder showing diffuse interfollicular infiltration with plasma cells and lymphocytes, as described in the preceding section (Fig. 7*c* and *d*). The thyroiditis was considered to be moderate and focal in 130 cases (Fig. 8), and it was listed as slight in 150 cases (Fig. 9). In the majority of the latter cases, focal collections of lymphocytes with lymph follicles formed a prominent part of the "thyroiditis," and the interfollicular infiltration usually was fairly minimal in amount.

The uninvolved parenchyma in these focal lesions also presented considerable variation. Many thyroids showed much colloid-containing tissue, the epithelium of which was normal or slightly swollen and oxyphilic. In others, macrofollicular and somewhat oxyphilic thyroidal tissue was present, with a greater or lesser degree of papillary infolding. The latter change sometimes produced a distinct microscopic parenchymal nodularity.

The possibility that thyroiditis of this type, with its diffuse interfollicular infiltration, may be superimposed on a previous colloid goiter was suggested in 17 patients; 16 of them were females and 1 was a male. The aver-

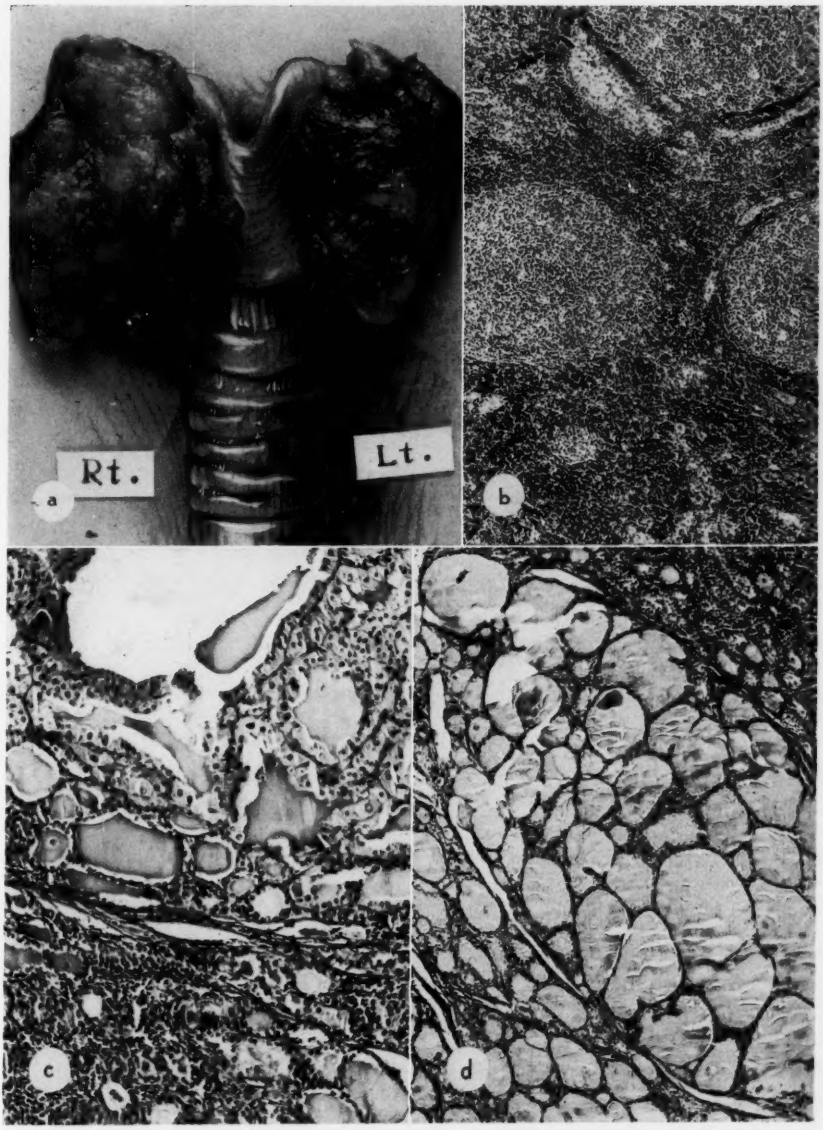

FIG. 8. Focal thyroiditis. **a.** Note moderate involvement. **b.** Same case, showing severe lymphocytic infiltration, with lymph follicles (×50). **c.** Same case, with focal areas of oxyphilic epithelium (×100). **d.** Same case, with areas of fairly normal uninvolved parenchyma (×60).

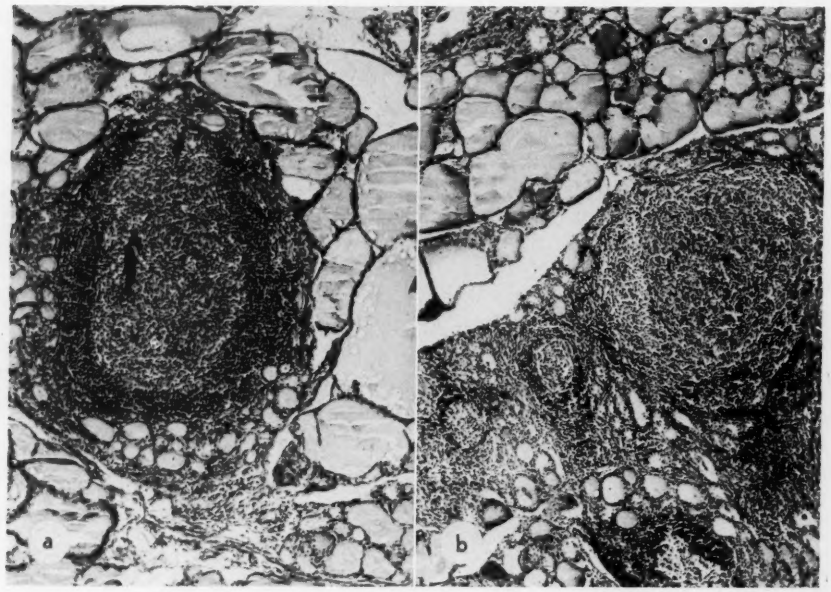

FIG. 9. Focal thyroiditis with only slight involvement. **a.** Lymphocytic aggregate with lymph follicles (×65). **b.** Another case, with larger focal lymphoid islands in a normal-appearing background (×60).

age duration of the goiter was fifteen years. All 17 patients were euthyroid. All the goiters were large, the average weight of resected tissue being 117 Gm. Histologically, the degree of thyroiditis was graded as slight and spotty in 4 cases and as moderate, frequently with diffusely involved zones, in 13. In 1 instance there was considerable morphologic evidence of progression from slight to pronounced involvement over a period of seven years. The patient, a 56-year-old euthyroid woman, had a goiter of fifteen years' duration. Operation in 1935 disclosed a large goiter with considerably more enlargement on the right. Subtotal resection of the right lobe only was performed. The resected tissue weighed 111 Gm. Histologically, a colloid type of goiter with minimal spotty thyroiditis was present (Fig. 10*a*, *b* and *c*). In 1942, the left lobe, which had been progressively enlarging, was removed. The resected tissue weighed 120 Gm. It showed diffuse thyroiditis with interfollicular plasma-cell and lymphocytic infiltration involving the bulk of the lobe, and zones of lesser involvement suggesting a colloid type of goiter, as in the preceding specimen (Fig. 10*d*, *e* and *f*).

A few thyroids presented evidence of nodularity of the hypertrophy-involution type, suggesting origin in a toxic or nontoxic nodular goiter. The

degree to which Graves' disease may be complicated by thyroiditis of this type cannot be assessed from this series, as already explained.

Many patients had some degree of swelling of cytoplasm and oxyphilia in the thyroidal epithelium. Suggestive evidence of considerable prolifera-

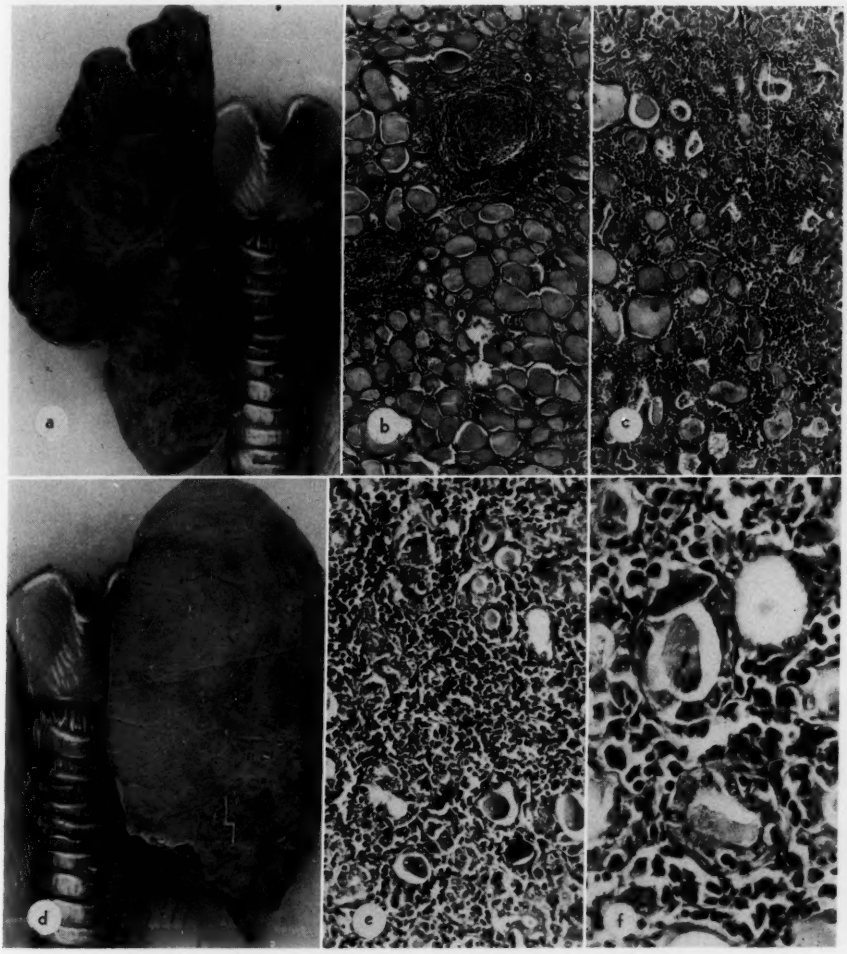

FIG. 10. Focal thyroiditis with subsequent transformation to diffuse type. **a, b** and **c.** Right lobe of thyroid removed in 1935. **a.** Note large amount of colloid-containing parenchyma. **b.** Section representative of most of gland, showing abundant colloid (reduced from ×60). **c.** Small zones of focal thyroiditis (reduced from ×85). **d, e** and **f.** Left lobe of thyroid in same case removed in 1942. **d.** Note extensive thyroiditis. **e.** Representative section, showing extensive infiltration (reduced from ×200). **f.** Higher-power view of same zone (reduced from ×415).

tive activity is sometimes observed in this type of epithelium, and a definite nodularity in the gland is seen when the change is extreme (Fig. 11). This change has been referred to as "regenerative hyperplasia," and it was sufficiently striking in 66 cases to warrant comment. In most instances, associated moderate spotty infiltration of lymphocytes and plasma cells was observed, although the amount of infiltration in a few was scanty, and the

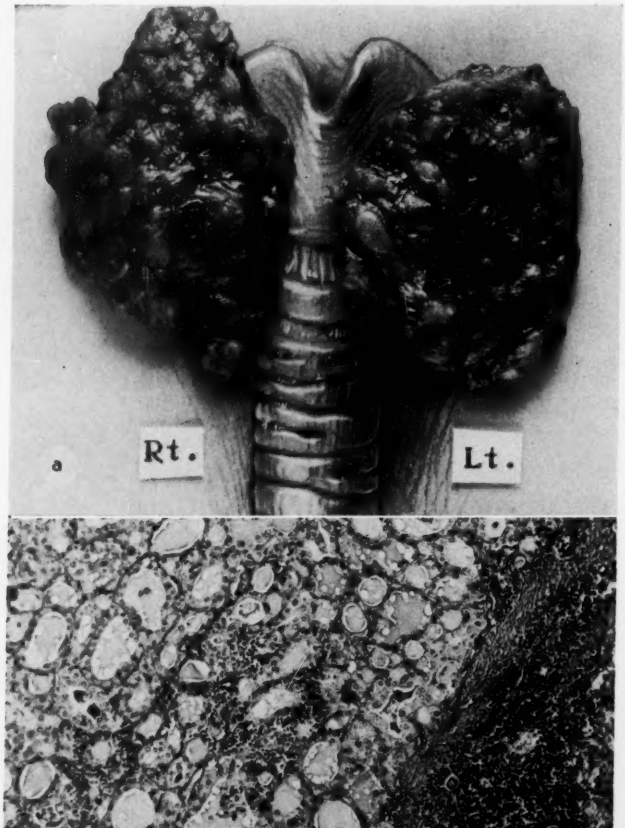

FIG. 11. Focal thyroiditis. **a.** Nodular gland with oxyphilic epithelium but minimal cellular infiltration. **b.** Portion of a nodule in another case, showing altered epithelium and scanty infiltration (×95).

epithelial changes were the dominant histologic feature (Fig. 11*b*).

Course. Bilateral subtotal resection was the surgical procedure in 314 patients, and follow-up data were available on 178 of them. Postoperative myxedema developed in 157 (88 per cent), whereas the remaining 21 were known to be euthyroid. Unilateral subtotal thyroidectomy was performed on 46 patients, with follow-up data available on 21; myxedema developed in 12 of these (57 per cent), and 9 remained euthyroid.

COMMENT

A number of features in this study are worthy of special comment.

Recurrence of goiter. Recurrence of goiter in struma lymphomatosa after surgical removal is not common, and pathologic data concerning the morphologic changes seen at intervals over a period of years are correspondingly difficult to obtain. Among the patients with diffuse involvement, goiter did not recur in those whose glands showed extensive epithelial destruction. Two patients with oxyphilic thyroidal epithelium had recurrent goiters. One of them had undergone thyroidectomy elsewhere three years prior to treatment at the clinic for a small recurrent lesion, and had been under treatment for myxedema when the recurrence took place. Comparison of the 2 specimens from this patient revealed an essentially similar degree of thyroiditis, with oxyphilia being slightly more pronounced in the second one. The other patient, who was euthyroid when seen at the clinic, had a huge recurrent goiter (172 Gm.) removed here; this patient had undergone previous thyroidectomies elsewhere one and five years previously. Sections from the earlier specimens were not available.

Recurrence was noted in 7 of the patients who had "varied epithelium," previous thyroidectomy having been performed eight to twenty-five years before. Tissue was not available in these cases. Only one lobe had been removed at the first operation in a number of these instances. One recurrence was noted in the hyperplastic group; this patient was euthyroid and had undergone thyroidectomy of unknown extent four years prior to operation at the clinic. Two recurrences have been referred to previously in this paper, 1 in the lymphoid group and 1 in the focal group; tissue was available from the 2 procedures in both instances. The former patient had myxedema with the recurrence, whereas the latter was euthyroid. Recurrence was noted in 11 additional patients of the group with "focal thyroiditis." Of these, 8 were euthyroid, 2 had myxedema and 1 was considered borderline in respect to myxedema.

Thus, relatively few pathologic data are available to document progressive morphologic changes over a period of years, although such progression is suggested by the gradual development of myxedema in many patients after biopsy or unilateral subtotal resection. That such progression does

not necessarily always take place is illustrated by the following case.

A 22-year-old euthyroid woman came to the clinic in 1922 with a history of goiter since the age of 14. Both lobes were enlarged to twice normal size. Bilateral subtotal thyroidectomy was performed, and an invasive papillary carcinoma 3 cm. in diameter was found in the left lobe. The remainder of the thyroidal tissue (30 Gm.) exhibited moderate thyroiditis with numerous lymph follicles (Fig. 12*a*). On her second admission, which was in 1936, she was euthyroid and had noted recurrence of goiter for six years. Operation was advised because of the size of the recurrent goiter. Bilateral subtotal resection again was performed; 40 Gm. of tissue was removed that histologically resembled the previously resected gland (Fig. 12*b*). In 1946, the patient noted a recurrence of six months' duration, and 50 Gm. of tissue was removed from the right side. Histologically, the tissue again showed spotty thyroiditis, perhaps slightly more pronounced than in the preceding specimens (Fig. 12*c*). The patient was last seen in 1956, with a soft recurrent goiter. She was euthyroid, the BMR was −3 per cent, and the thyroidal uptake of I^{131} was 15 per cent in twenty-four hours.

Morphologic findings and definition. It would appear that Hashimoto's thyroiditis is characterized mainly, in its advanced form, by diffuse interfollicular infiltration with plasma cells and lymphocytes. The associated epithelial changes suggest varying degrees of follicular damage. Spotty or even total destruction of epithelium is present in some cases.

Oxyphilic epithelium is a common finding, presenting a striking histologic picture in its advanced diffuse form. A number of clinical and laboratory findings, including a low average basal metabolic rate, the occurrence of preoperative myxedema in 25 per cent of this particular subgroup, and low-normal or subnormal levels of serum protein-bound iodine, combined with a normal or even increased thyroidal uptake of I^{131}, suggest a limited degree of functional activity for this oxyphilic type of cell. On the other hand, a number of patients without oxyphilia but with diffuse infiltration had myxedema and similar functional changes. It is a reasonable deduction from the histologic findings that patients with total or almost total destruction of epithelium should show almost total lack of thyroidal function, as outlined. The duration of the goiter was, if anything, somewhat shorter in this group, strongly suggesting a variation in tempo or intensity of the destructive process in some patients.

There would not appear to be sufficient difference between the clinicopathologic findings in the 4 subgroups of diffuse thyroiditis described in this paper to warrant arbitrary selection of any one as representing true "Hashimoto's thyroiditis," and it appears reasonable to consider the group as a whole as representing an advanced stage of the disease process, be it auto-immunization or something else.

The question as to whether glands with focal thyroiditis should be placed in a separate category is a more difficult one to decide with the data at hand. Many of these thyroids, on morphologic evidence, strongly sug-

gest various stages in the development of the disease and, when this is fairly pronounced, they differ but little from glands with more advanced changes. Until this question is settled, it is evident that statistics concern-

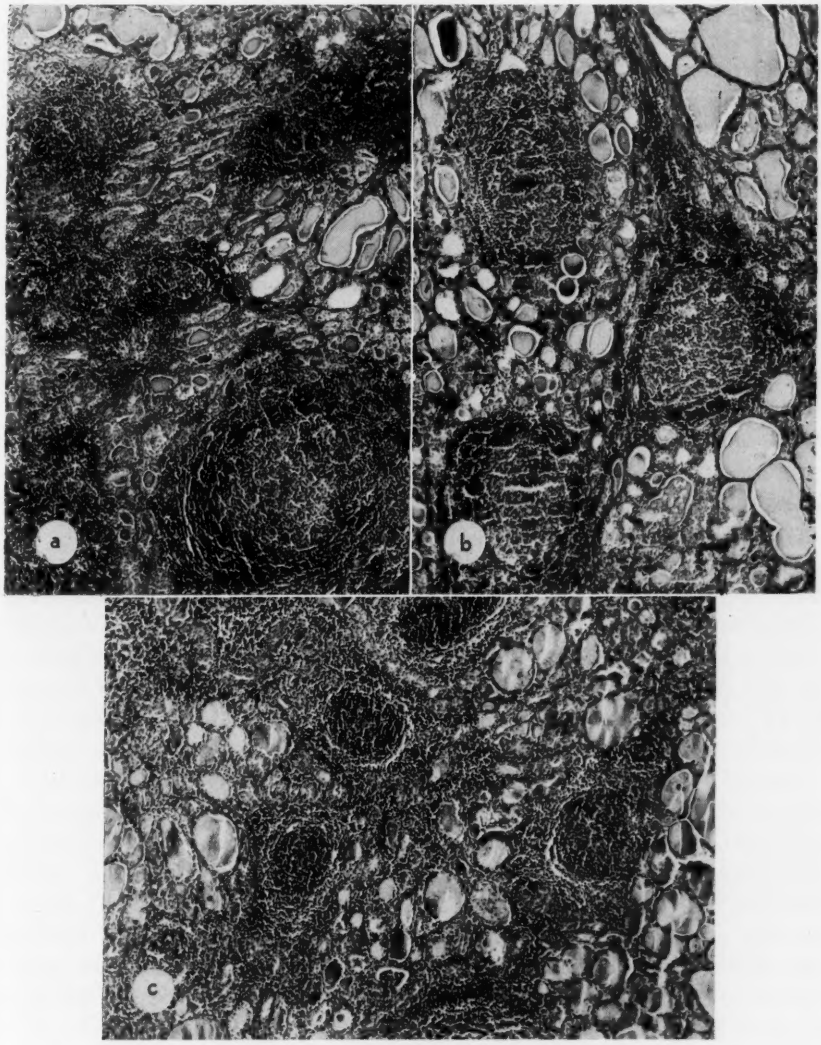

FIG. 12. Focal thyroiditis, with recurrence of goiter. **a.** Representative section from thyroid removed in 1922 (×75). **b.** Recurrent lesion removed in 1936. There is an approximately equal degree of involvement (×65). **c.** Recurrent lesion removed in 1946. The over-all histologic pattern is closely similar to that seen in **a** and **b** (×65).

ing many aspects of "Hashimoto's disease" will tend to vary according to whether such cases have been included or excluded.

In regard to these cases in which the involvement is focal, one is impressed by the need for further information on whether progression of the pathologic process seldom, usually or always takes place, and what variation in tempo and degree of morphologic and functional alteration may be expected in such instances. Also, further study of the "soil" in which thyroiditis develops appears necessary. This is, perhaps, more readily accomplished by a consideration of the cases in which varying degrees of focal change are present rather than cases characterized by far-advanced lesions. The impression has been gained from this study that a variety of thyroidal states, either normal or pathologic, may be complicated by thyroiditis of this type. Clinical data in the cases in this series, however, suggest that certain conditions such as thyrotoxicosis and granulomatous thyroiditis are only infrequently the precursors of the fully advanced classic disease.

The group of patients in this series designated as having "hyperplastic" changes is likewise worthy of further study. This type of morphologic disturbance is the common one in younger patients, the average age in this series being 31 years, which was sixteen to twenty-three years less than the averages in the other "types." This group apparently corresponds to those cases reported by Gribetz and associates (7) as representing "lymphocytic thyroiditis" in preadolescent and adolescent girls. In our series, the sex incidence was overwhelmingly female (56:1). Almost half of our patients were in the first 3 decades of life, but some older patients showed essentially similar morphologic findings. Gribetz and co-workers found abnormally high values for serum protein-bound iodine concentration in 3 of 6 children in their series. Studies of thyroidal function in our patients showed normal levels of serum protein-bound iodine in 5 of 8 patients in the group, 2 of these being "high normal"; the value was less than normal in the other 3 (Table 7). Gribetz and associates (7) and, later, Owen and McConahey (8) stated that the discrepancy often found in patients with Hashimoto's thyroiditis between the values for protein-bound iodine and butanol-extractable iodine may be due to a non-calorigenic or low-calorigenic "thyroglobulin" in the serum. This impairment in the production of thyroxine is further emphasized by observations in a small number of cases of this type seen since the completion of this study in which normal values for serum protein-bound iodine were associated with greatly decreased levels of butanol-extractable iodine. Radioiodine studies on 14 patients (Table 7) indicated that these thyroids were collecting iodine well, although the production of thyroxine was impaired.

Association with carcinoma or malignant lymphoma. An associated carcinoma was present in 18 of these 605 cases of "thyroiditis" seen over a 27-

year period. These carcinomas may be arbitrarily divided into tumors of ordinary size (1.5 to 4 cm. in diameter) and small tumors of approximately 1 cm. or less in diameter. Of the 18 carcinomas, 7 were of ordinary size, 2 being associated with involved lymph nodes; the remaining 11 were small carcinomas, 1 of which was multiple. Eight of the latter were discovered on careful re-examination of multiple blocks of the thyroid as outlined. All these carcinomas, both large and small, were of papillary type and of a low grade of malignancy. None was included unless definite invasion of adjacent thyroidal parenchyma could be demonstrated. The significance of small papillary carcinomas, which appear to be relatively common in any type of thyroid, including those affected by thyroiditis, requires further study.

Lymphosarcoma was noted 12 times in the 605 cases but, as already explained, certain cases of lymphosarcoma were not included in this study, since knowledge of the adjacent parenchyma could not be obtained. In only 3 cases was diffuse thyroiditis present in the extratumorous tissue. The process was focal and sometimes minimal in degree in the remainder, consisting mainly of scattered islands of lymphocytes with germinal centers.

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CLINICAL OBSERVATIONS ON SERUM GLOBULIN THYROXINE-BINDING CAPACITY, USING A SIMPLIFIED TECHNIQUE*

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ABSTRACT

Using a relative simplified and reproducible technique, the thyroxine-binding capacity of the serum was measured in normal subjects, in pregnant women, in cases of hepatic cirrhosis, hypothyroidism and hyperthyroidism, and in euthyroid subjects with subnormal and elevated levels of serum protein-bound iodine (PBI).

In 103 normal subjects having serum PBI levels between 5 and 7 μg. per 100 ml., the mean value of the thyroxine-binding capacity was 19.1 μg. per 100 ml.; female subjects were not significantly different from male subjects in this respect. In 34 men with low PBI values (between 2 and 4 μg. per 100 ml.), the mean was 13.9 μg. per 100 ml.

In 33 pregnant women, both the thyroxine binding capacity and the PBI concentration were increased. They returned to normal or lower levels by the end of the fifth week post partum. Although the thyroxine-binding capacity was often slightly higher than normal in 20 patients with hepatic cirrhosis, no definite correlation was found between the thyroxine-binding capacity, PBI level and the presence of vascular spiders.

The binding capacity was slightly elevated in 9 patients with myxedema, whereas it was normal or lower than normal in 13 hyperthyroid patients. In 10 of 14 female euthyroid subjects with abnormally high serum PBI values, the thyroxine-binding capacity was definitely elevated. The question of the physiologic importance of thyroxine-binding globulin is discussed.

INTRODUCTION

IT IS well known that normally most of the thyroxine in blood is bound by a specific globulin (TBG) which has an electrophoretic mobility intermediate between alpha-1 and alpha-2 globulin at pH 8.6 (1–4). The remainder of the thyroxine is attached to the albumin fraction.

Several observations in pregnancy (5–6) and nephrosis (7) seem to indicate that the peripheral action of the thyroid hormone is a function of the free thyroxine in the blood, and that this level is controlled by the binding

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capacity of TBG. Whether or not this is so, it would seem that the level of TBG should be taken into consideration in the interpretation of serum PBI values.

Since it is not possible to determine the absolute amount of TBG in the plasma directly, the thyroxine-binding capacity of this protein has been used as an index of its concentration.

A reliable method has been developed for the determination of the binding capacity of TBG. This capacity has been measured in normal subjects, in pregnant women, in cases of hepatic disease, hypothyroidism and hyperthyroidism, and in euthyroid subjects with subnormal and elevated levels of serum protein-bound iodine (PBI).

METHODS

Thyroxine-binding capacity was estimated by paper electrophoresis of the serum to which had been added a relatively great amount of radioactive thyroxine. Chromatographically pure radioactive thyroxine[1] in 50 per cent propylene-glycol was diluted with more propylene-glycol to make a solution containing approximately 10 micrograms of thyroxine per milliliter. Of this solution, 0.05 ml. was placed in a small flask and evaporated to dryness by a vacuum pump at room temperature; 0.5 ml. of serum was added to the residue. The final concentration of thyroxine in this mixture was calculated from the iodine content (chemically determined) of the serum and of the original thyroxine solution, and by recovery of radioactivity to correct for losses of thyroxine on glassware or during evaporation.

The thyroxine-serum mixture was studied by paper electrophoresis in veronal buffer (pH 8.6, ionic strength 0.1) employing the reverse-flow method of Robbins (8). After making a radioautogram from the electrophoresed papers, they were placed on the autogram, and cut along the 2 darkened areas corresponding to albumin and TBG. The radioactivity of these papers was counted in a well-type scintillation counter.

The absolute amount of thyroxine bound to TBG can be ascertained by multiplying the percentage of radioactive thyroxine in the TBG by the concentration of thyroxine in the serum. This reaches a plateau at thyroxine concentrations above approximately 50 μg. per 100 ml. in normal serum (Fig. 1). Even in serum with an increased binding capacity, 100 μg. of thyroxine per 100 ml. of serum was sufficient for the saturation of TBG.

Reproducibility: Two specimens of serum, each divided into 10 aliquots, were electrophoresed and analyzed. The standard deviation for each 10 determinations was less than 6 per cent of the mean value (Table 1).

RESULTS

Normal values

One hundred and three normal subjects with serum PBI levels between 5 and 7 ug. per 100 ml. were divided into 4 groups according to the age and sex. No significant difference in the TBC of the serum was found among

[1] Obtained from Abbott Laboratories, Oak Ridge, Tennessee.

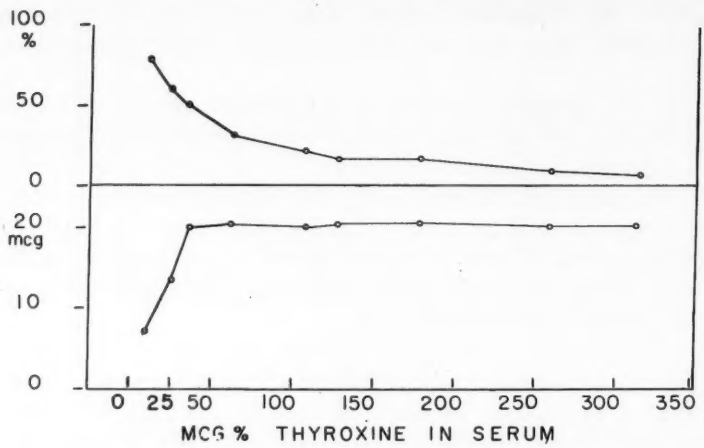

FIG. 1. The upper graph shows the percentage of total radioactivity, and the lower graph shows the amount of thyroxine in TBG at various concentrations of serum thyroxine (exogenous plus endogenous) expressed in μg. per 100 ml.

these groups. The ranges, means and standard deviations of the thyroxine-binding capacity in 61 males and 42 females are summarized in Table 2.

Males with low serum PBI level

There were 33 males with a low level of the serum PBI, ranging from 2.4 to 4.0 ug. per 100 ml. In these men the TBG was characteristically lower than in the men with normal serum PBI values; the range was 9.5 to 21.7 μg. per 100 ml., with an average of 13.9 μg. If the low level of TBG were to allow a greater availability of free thyroxine, these subjects should be euthyroid, yet half had suggestive evidence of hypothyroidism, as indicated by hypercholesterolemia, low basal metabolic rate, increased percentage of serum beta lipoprotein, or low thyroidal I^{131} uptake. Their clinical condition raises serious doubt that a low TBG level promotes euthy-

TABLE 1. THE REPRODUCIBILITY OF DETERMINATIONS OF THYROXINE-BINDING CAPACITY IN 10 ALIQUOTS FROM TWO DIFFERENT SERUMS

Serum	TBC (μg./100 ml.)	
	Range	Mean ± S.D.
A	12.7–15.9	14.0±0.9
B	27.1–30.9	29.6±1.1

TABLE 2. SERUM PBI CONCENTRATION AND THYROXINE-BINDING CAPACITY OF TBG IN EUTHYROID SUBJECTS WITH NORMAL AND ABNORMAL PBI LEVELS; IN MYXEDEMA, HYPERTHYROIDISM AND HEPATIC CIRRHOSIS; AND PREGNANT WOMEN

	Sex	No. of subjects	PBI (μg./100 ml.)	TBC (μg./100 ml.)	
				Range	Mean ± S.D.
Euthyroid	♂	34	2–4	8.7–21.4	13.9± 2.6
	♂	61	5–7	11.3–30.0	19.1± 4.5
	♀	42	5–7	11.0–30.4	20.3± 4.9
	♀	14	8–24.2	16.0–49.3	34.4±10.0
Myxedema	♂ ♀	9	0.8– 3.4	14.7–43.6	26.4± 8.5
Hyperthyroidism	♂ ♀	13	8.1–22.8	8.8–24.9	17.8± 5.1
Hepatic cirrhosis	♂ ♀	20	3.7–10.6	12.2–38.0	24.6± 7.3
Pregnancy*	♀	33	4.1–10.4	21.7–62.3	41.3±10.1

* After 12 weeks pregnancy.

roidism in spite of a low serum thyroxine concentration (PBI). For example, A.L., a 23-year-old medical student, on one occasion had a serum PBI level of 2.4 μg. per 100 ml.; his TBG level was only 9.8 μg. per 100 ml.; yet his BMR was −19 per cent, his thyroidal I^{131} uptake was 10.9 per cent and he was 30 pounds overweight. On the contrary H.F.R., 48 years old, (not included in the TBG summary) had a PBI level of 3.5 μg. per 100 ml. and a TBG level of 41.4 μg. per 100 ml. He should have been hypothyroid because of the greatly reduced availability of free thyroxine; yet his BMR was −9 per cent, the thyroidal I^{131} uptake was 14 per cent, and the serum cholesterol level was 215 mg. per 100 ml.

Pregnancy

The thyroxine-binding capacity as well as the serum PBI level began to increase in early pregnancy and remained elevated during the whole course. The magnitude of the increase in the thyroxine-binding capacity was much greater than that of the elevation in the PBI concentration.

After delivery of the babies, both serum levels started to decrease within a few days and were definitely diminished in the second week post partum. In 8 out of 9 cases, both values returned to normal or lower levels by the end of the fifth week (Table 2 and Fig. 2).

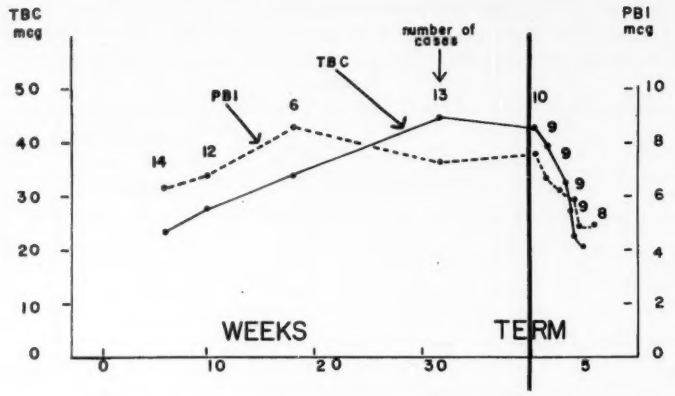

FIG. 2. Changes in thyroxine-binding capacity and serum PBI level during pregnancy and the puerperium.

Points are averages for different groups. Solid line = thyroxine-binding capacity of TBG (μg. per 100 ml.). Dotted line = concentration of serum PBI (μg. 100 ml.).

Hepatic cirrhosis

The thyroxine-binding capacity in 20 patients with hepatic cirrhosis was slightly augmented—mean value, 24.4 μg. per 100 ml. (Table 2). There was no statistical difference in binding capacity or PBI level between patients with vascular spiders and those without (Fig. 3).

Hypothyroidism and hyperthyroidism

Prior to treatment, the thyroxine-binding capacity in 9 cases of myxe-

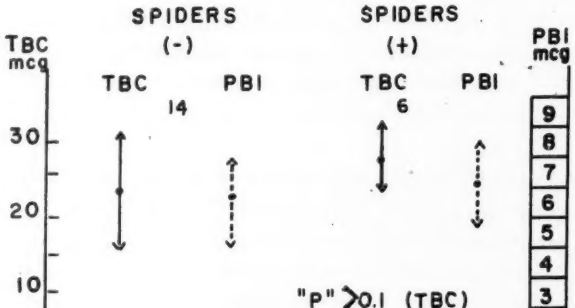

FIG. 3. The thyroxine-binding capacity (μg. per 100 ml.) and serum PBI level (μg. per 100 ml.) in cirrhotic patients with and without vascular spiders.

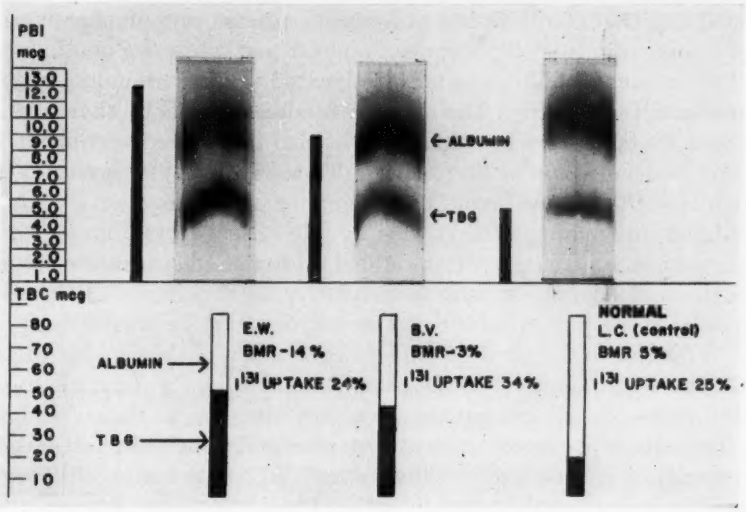

FIG. 4. Electrophoresis of serum from 2 euthyroid subjects with elevated serum PBI levels and from 1 normal subject.

In the upper half of the figure, the narrow black columns indicate the concentration of serum PBI (μg. per 100 ml.); the wide columns represent radioautograms made from the electrophoresed papers, with 2 dark areas in each corresponding to albumin and TBG respectively. In the lower half of the figure, the columns represent the thyroxine-binding capacity (μg. per 100 ml.) as distributed between albumin and TBG.

dema was slightly higher than normal, whereas that of 13 hyperthyroid patients was in the normal range (Table 2).

During treatment of the hypothyroid patients, the abnormal thyroxine-binding capacity had a tendency to revert towards normal values. However, no conclusion can be drawn at this time, since serial studies in individual cases were not made.

Euthyroidism with elevated serum PBI level

Fourteen euthyroid subjects with elevated serum levels of PBI and butanol-extractable iodine were studied. Their basal metabolic rates and thyroidal I^{131} uptakes were normal. They showed no evidence of hyperthyroidism. In 10 of the 14 cases, the serum thyroxine-binding capacity was increased (Table 2). Figure 4 illustrates the radioautograms in 2 subjects with elevated levels of PBI and thyroxine-binding capacity and in 1 normal control subject.

DISCUSSION

Gordon and associates (1) first demonstrated in 1952, that the thyroxine

in blood is preferentially bound to a specific plasma protein characterized by electrophoretic mobility between alpha-1 and alpha-2 globulin at pH 8.6. This specific globulin is usually designated thyroxine-binding globulin or protein (TBG or TBP). The selective binding of TBG by thyroxine has also been shown *in vitro* by adding I^{131}-labeled thyroxine to serum (9–10).

As the concentration of thyroxine added to serum is increased, the proportion of the thyroxine bound to albumin increases progressively. On the other hand, the binding of thyroxine by TBG has a certain limit. TBG is usually saturated *in vitro* when the added thyroxine concentration is two or three times that of the thyroxine circulating *in vivo*. Since TBG has not been isolated in a pure form nor determined directly, the maximal capacity of TBG for thyroxine has been studied as an index of the concentration of TBG. The technique used in the present studies for the determination of the thyroxine-binding capacity is essentially the same as that of Robbins; good reproducibility seems to have been obtained using radioautograms.

Although there is no direct evidence that TBG participates in the regulation of the peripheral distribution of the thyroid hormone, several investigators (5–7, 11) have suggested that there is a dynamic equilibrium bebetween the thyroxine bound to TBG and the free thyroxine in the blood which is available to peripheral tissues.

Accurate basal metabolic rate studies of this relationship, however, have not been made. Several findings contradictory to this theory appear in this study: 1) In the 33 males with a low level of serum PBI, the very low thyroxine-binding capacity did not prevent hypothyroidism. 2) A patient with no TBG, and a serum PBI level of 3.0 μg. per 100 ml. was found; other data indicated that he was probably euthyroid. 3) In pregnancy, the rise of the serum PBI level was not maintained in proportion to the increase in TBG, and 4) In 4 of the euthyroid patients with elevated serum PBI levels, the concentration of TBG was normal.

Alterations in the quality of binding of thyroxine to this globulin should be considered.

Apparently globulin is not an obligatory transport protein for thyroxine. Certainly triiodothyronine reaches the tissues through the circulation without accumulating sufficiently to be measurable by the clinical serum PBI procedure.

Further studies of thyroxine-binding globulin are necessary to clarify its physiologic role.

Acknowledgment

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THYROID FUNCTION IN SUPRAVENTRICULAR TACHYCARDIAS: TURNOVER OF INTRAVENOUSLY INFUSED I^{131}-LABELED THYROXINE AND THE RED BLOOD CELL UPTAKE OF I^{131}-LABELED *l*-TRIIODOTHYRONINE*†

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ABSTRACT

Thyroid function was studied in 28 euthyroid patients with paroxysmal or recent prolonged atrial fibrillation and in 11 patients with paroxysmal atrial tachycardia. The serum protein-bound iodine level, basal metabolic rate, and thyroidal I^{131} uptake were within normal limits. In 11 of these cases, the fate of intravenously administered I^{131}-thyroxine was studied. The half-time of disappearance of plasma radioactivity following infusion of I^{131}-labeled thyroxine ranged from 4.1 to 6.8 days and averaged 5.74 days—significantly faster than the turnover in previous studies of euthyroid human subjects. The thyroxine distribution space ranged from 8.0 to 16.4 liters (average, 12.2 liters), in contrast to a range of 7 to 12 liters (average, 9.9 liters) in the control group. The daily degradation rate of thyroxine varied from 52 to 126 micrograms per day (average, 83.6) in the group under study but was only 36 to 61 micrograms in the control group. The two-hour *in vitro* red blood cell uptake of I^{131}-triiodothyronine from whole blood in 18 male patients with atrial arrhythmias was 18.6 per cent per 100 hematocrit. In 8 of these cases, the values were in the range often seen in thyrotoxicosis. The red blood cell uptake of I^{131}-triiodothyronine in 21 female patients similarly studied averaged 16.5 per cent. Seven of the 21 had uptakes greater than 18 per cent, the upper limit for euthyroid females. In the 28 patients with atrial fibrillation, the red blood cell uptake was abnormal in 12. In the 11 patients with paroxysmal atrial tachycardia, the red blood cell uptake was abnormal in 3. Five patients were treated with large doses of potassium iodide, although they were considered euthyroid. Significant improvement was noted in each.

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THE relation of the thyroid gland to cardiac function and rhythmicity has long been recognized. In 1825, there were recorded, posthumously, observations made by Parry in 1786 of patients with a malady in which enlargement of the thyroid gland accompanied palpitation of the heart and tachycardia (1). Clinical confirmation has been readily gained from reports on patients with thyrotoxicosis accompanied by sinus tachycardia, atrial fibrillation, atrial flutter or atrial tachycardia (2, 3).

Observations in our laboratory have suggested an influence of the thyroid on cardiac irritability in other instances. Sixteen euthyroid patients with rheumatic heart disease, congestive heart failure and long-standing atrial fibrillation were treated by the induction of hypothyroidism with I^{131}. Reversion to sinus rhythm occurred in 3 of the 16 patients following the induction of hypothyroidism. Furthermore, when thyroid function was lowered with I^{131} in euthyroid patients with frequent episodes of disabling paroxysmal fibrillation, the attacks became less frequent and less severe (4a and b). These additional indications of the close relationship between thyroid function and cardiac rhythm led us to investigate thyroid function in euthyroid patients with supraventricular arrhythmias.

MATERIALS AND METHODS

Thyroid function was studied in 39 patients with supraventricular arrhythmias. Twenty-eight patients, aged 40 to 80, had atrial fibrillation; in 24 it was paroxysmal and in 4 it was of recent onset (having failed to revert to sinus rhythm within a month). Ideally, in the diagnosis of paroxysmal atrial fibrillation, the following findings should be available : 1) a normal electrocardiogram prior to the episode, 2) an electrocardiogram showing fibrillation during the paroxysm, and 3) a normal electrocardiogram following the attack. Such complete data are rarely obtained. In the cases presented here, the diagnosis was verified either by electrocardiogram or by an examination by a competent cardiologist. Ten of these patients were free of obvious heart disease. However, since 8 of them were males between 45 and 60 years of age, the probability of underlying coronary atherosclerosis was high. Seven of the 28 suffered from rheumatic heart disease, and the remaining 11 from hypertensive or atherosclerotic cardiovascular disease. There were no known causes for the precipitation of the arrhythmia, although 1 patient had a low-grade fever and 1 had had a recent respiratory infection.

Eleven patients, aged 20 to 71, suffered from paroxysms of atrial tachycardia. In 6, the arrhythmias were documented electrocardiographically; in the others, there was a typical history of bouts of rapid regular heart action, sudden in onset, abruptly concluded. All were free of overt heart disease.

All 39 patients were judged to be euthyroid at the time of final evaluation after consideration of the clinical picture and appropriate laboratory tests (Tables 1 and 2). Serum protein-bound iodine (PBI) determinations were performed in every case in which thyroxine was infused or in which the red blood cell uptake was elevated (5). The serum protein-bound iodine level, measured in 29 of the 39 cases, ranged from 4.2 to 7.0 μg. per 100 ml.; in no case was it elevated. In all instances in which thyroidal I^{131} uptake was measured, it was within the euthyroid range for this laboratory (6). The basal metabolism (7) was measured in 19 cases and found to be normal in 18. It was elevated

TABLE 1. THYROID FUNCTION IN 11 PATIENTS INFUSED WITH I^{131}-THYROXINE*

Patient		$T\frac{1}{2}$ (days)	K (%/day)	TDS (liters)	D (μg./day)
No.	Wt. (lbs.)				
2	215	5.8	12.0	16.4	126
3	122	5.8	12.0	10.5	52
5	127	6.0	10.8	10.1	52
6	192	5.0	13.8	15.4	123
7	130	6.6	10.5	10.0	63
17	186	5.8	12.0	15.1	110
18	145	5.9	11.7	10.4	60
29	122	5.7	12.2	8.0	62
31	181	6.8	10.3	16.1	88
32	197	4.1	16.9	10.8	105
M†	154	5.7	12.2	11,2	79
Average		5.7	12.2	12.2	83.6
SD		0.52			28.3

* $T\text{-}\frac{1}{2}$ = Half-time of disappearance of I^{131}-thyroxine.
K = Turn-over rate for thyroxine.
TDS = Thyroxine distribution space.
D = Degradation rate for thyroxine.

† Previous prolonged administration of estrogens. Not included in Table 2. Serum PBI level = 5.8 μg. per 100 ml.; thyroidal I^{131} uptake = 30%; r.b.c. uptake = 10.5%.

(+10 per cent or more) in 1 patient; she was extremely apprehensive in the presence of disturbing atrial fibrillation, and it was felt that the metabolic elevation reflected apprehension rather than hyperthyroidism; the serum PBI level and thyroidal I^{131} uptake were normal. In all cases, the two-hour *in vitro* uptake of I^{131}-labeled triiodothyronine by the patient's red blood cells (from his own whole blood) was measured by the technique previously developed in this laboratory (8). In 11 patients, I^{131}-thyroxine was infused intravenously, and the thyroidal, urinary and plasma radioactivity was measured daily for seven to ten days as described elsewhere (9). From these data, the half-time of disappearance of thyroxine, the percentage turned over daily, the volume of distribution of thyroxine and the micrograms of thyroxine degraded daily were calculated (10).

RESULTS

The effects of infusion of I^{131}-thyroxine are shown in Table 1. The half-time of disappearance of plasma radioactivity ranged from 4.1 to 6.8 days and averaged 5.7 days (S.D., 0.52; S.E., 0.16). In our previously reported series of normal subjects, the half-time of disappearance of radioactivity following infusion of I^{131}-thyroxine ranged from 6.6 to 7.8 days and averaged 7.2 days (S.D., 0.43; S.E., 0.13). The difference between these means is significant at the level $P < .001$. The mean daily proportion of thyroxine turned over in the patients with arrhythmia was 12.2 per cent, and in the

TABLE 2. THYROID FUNCTION IN ATRIAL FIBRILLATION AND PAROXYSMAL ATRIAL TACHYCARDIA

Patient		Diagnosis*	RBC (%/100 het.)	Serum PBI (μg./100 ml.)	BMR (%)	Thyroidal I^{131} uptake (%)
No. & Initial	Sex & Age					
1. S	M, 69	AF; CAD	25.5	5.8	+ 1	
2. K	M, 57	AF; OHD	19.5	4.8	− 5	8
3. S	F, 40	PAF; OHD	16.8	4.2	− 6	25
4. S	M, 80	PAF (old); CVA; OHD	20.6	6.9	—	—
5. F	F, 61	AF; RHD	16.7	4.8	+ 8	—
6. H	M, 47	PAF; OHD	16.3	5.8	− 6	39
7. McD	F, 64	AF; CAD	16.3	6.0	+ 7	—
8. B	M, 60	PAF; OHD	20.3	4.6	—	—
9. S	M, 50	PAF; RHD	24.7	6.2	—	17
10. K	M, 60	PAF; OHD	26.6	4.2	−15	—
11. N	F, 45	PAF; RHD	14.0	4.8	− 3	—
12. T	M, 66	PAF; OHD	13.8	4.6	—	—
13. A	F, 45	PAF; RHD	12.5	5.2	—	—
14. S	F, 62	PAF; PAT; HHD	21.5	—	—	—
15. J	F, 75	PAF; CAD	20.9	4.6	—	31
16. M	F, 55	PAF; HHD	13.1	—	—	—
17. R	M, 50	PAF; OHD	17.5	7.0	− 7	37.7
18. G	F, 57	AF; HHD	18.8	5.9	+20	—
19. W	M, 54	PAT; PAT; CAD	19.5	7.0	+ 3	24
20. A	F, 40	PAF; OHD	18.9	6.4	+ 4	21
21. S	F, 56	PAF; RHD	16.0	6.8	—	12
22. H	M, 44	PAF; ?CAD	18.3	4.2	—	—
23. K	F, 42	PAF; RHD	17.1	4.0	—	32
24. G	F, 62	PAF; CAD	16.6	5.0	—	—
25. D	M, 48	PAF; OHD	18.6	—	—	—
26. C	F, 71	PAF; RHD; ?CAD	12.2	—	—	—
27. B	M, 45	PAF; CAD	13.6	—	—	—
28. G	M, 60	PAF; CAD	16.6	—	—	—
29. L	F, 39	PAT; OHD	15.5	6.4	−19	—
30. W	F, 38	PAT; OHD	21.5	5.2	+ 2	—
31. K	M, 60	PAT; OHD	20.2	5.3	+ 4	27
32. F	M, 71	PAT; OHD	14.0	5.8	−20	—
33. W	F, 45	PAT; OHD	14.0	—	−14	—
34. R	F, 20	PAT; OHD	14.0	—	—	—
35. S	M, 35	PAT; OHD	14.0	5.4	− 6	32
36. F	M, 60	PAT; OHD	15.0	—	—	—
37. B	F, 33	PAT; OHD	15.1	—	—	—
38. K	F, 65	PAT; OHD	13.3	—	+ 2	13
39. E	F, 55	PAT; OHD	19.5	—	—	—

* AF = Atrial fibrillation of more than one month's duration.
PAF = Paroxysmal atrial fibrillation.
PAT = Paroxysmal atrial tachycardia.
CAD = Coronary artery disease.
RHD = Rheumatic heart disease.
HHD = Hypertensive cardiovascular disease.
OHD = No overt heart disease.

normal subjects 9.7 per cent. The thyroxine distribution space in the patients with tachycardia ranged from 8.0 to 16.4 liters (average, 12.2 liters) in contrast to a range of 7 to 12 liters (average, 9.9 liters) in the control group. The daily thyroxine degradation varied from 52 to 126 micrograms per day (average, 83.6; S.D., 28.3; S.E., 8) in the group under study but was only 36 to 61 micrograms (average, 50.9; S.D., 8.6) in the control group. The difference between these means is significant at the level $P < .01$.

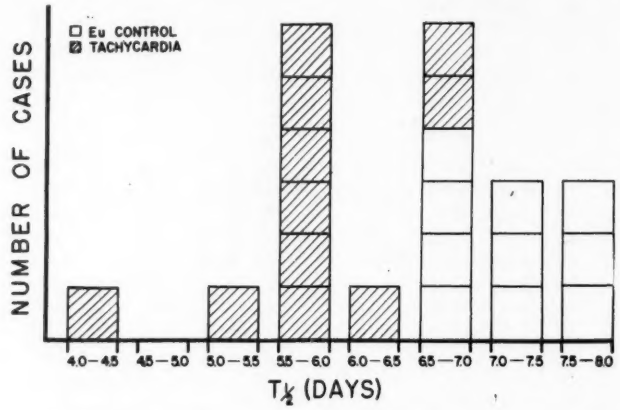

FIG. 1. Half-life of I^{131}-labeled *l*-thyroxine after intravenous infusion.

The two-hour *in vitro* red blood cell (r.b.c.) uptake of I^{131}-triiodothyronine from whole blood in the entire series of 39 patients averaged 17.5 per cent (Table 2). Because of the observed difference between values in euthyroid male and female subjects, we separated our results according to the sex of the patients (11). In 18 male patients with atrial arrhythmias, the r.b.c. uptake averaged 18.6 per cent/100 hematocrit. The average value for euthyroid male subjects is 15.2 per cent. Only 4.1 per cent of normal male patients have uptakes greater than 19.0 per cent. On the other hand, 8 of the 18 male patients with arrhythmias, although euthyroid, had r.b.c. uptakes greater than 19.5 per cent—values often associated with thyrotoxicosis. Twenty-one female patients with atrial fibrillation and atrial tachycardia were similarly studied. The r.b.c. uptake of I^{131}-triiodothyronine averaged 16.5 per cent, in contrast to the average in normal females of 13.9 per cent. Seven of the 21 had uptakes above 18 per cent (considered by us to be the upper limit for euthyroid females).

In 28 of the 39 patients, the arrhythmia could reliably be diagnosed as atrial fibrillation. These patients were evenly divided as to sex. The average r.b.c. uptake was 18.0 per cent. Twelve of the 28 determinations were in

the abnormal range. Eleven patients, 7 female and 4 male, suffered from episodes of proxysmal atrial tachycardia. The average r.b.c. uptake in this group was 16.1 per cent. Only 3 of the 11 determinations were clearly in the abnormal range. There was no relation between the r.b.c. uptake of I^{131}-triiodothyronine and the rate of turnover of I^{131}-thyroxine.

Many of the patients with repeated bouts of paroxysmal fibrillation were strongly suspected of being thyrotoxic; but in each instance, the serum PBI level, the 24-hour thyroidal I^{131} uptake, and usually the basal metabolic rate, were within the euthyroid range. Five patients, 4 with frequent uncontrollable paroxysms of atrial fibrillation and 1 with recent prolonged atrial fibrillation, were treated with large doses of potassium iodide despite the fact that they were considered euthyroid. In each case, improvement resulted. In 4 of the 5, paroxysms of fibrillation became much less frequent; in the fifth, chronic fibrillation continued but at a slower rate and the patient was much more comfortable. In 3, the cessation of attacks coincided with a decrease in the r.b.c. uptake of I^{131}-triiodothyronine. The following case is illustrative:

Case 1. R.A., a 40-year-old woman, had suffered for nine years from bouts of rapid irregular heart beat. A diagnosis of atrial fibrillation had been made, and she was told repeatedly there was no evidence of underlying heart disease. Smoking was omitted and quinidine was prescribed briefly without success. Fourteen years previously, the basal metabolic rate had been −22 per cent. Desiccated thyroid was prescribed in doses up to 2 grains daily, but was omitted because of palpitation. There was no other history suggestive of endocrinopathy. On examination, the skin was cool and dry. The pulse was 120 initially but with rest gradually fell to 80. Blood pressure was 116/76 mm. Hg. The thyroid was barely palpable. Blood and urine were normal. The first basal metabolic rate was +13 per cent but on repetition it was +4 per cent. The serum PBI level was 6.4 micrograms per 100 ml. The 24-hour thyroidal I^{131} uptake was 32 per cent. The two-hour r.b.c. uptake of I^{131}-triiodothyronine was 18.9 per cent per 100 hematocrit. The patient was considered euthyroid. Saturated solution of potassium iodide was prescribed initially in a dosage of 10 drops twice a day and later three times a day. After nine days, the patient experienced a remarkable freedom from palpitation, despite effort which in the past had invariably evoked paroxysms. The r.b.c. uptake decreased to 12.5 per cent. She gradually reduced the dose of iodide, and there was a recurrence of palpitation. The r.b.c. uptake rose to 18.8 per cent. The dosage of iodide was increased to 1800 mg. daily in enteric-coated tablets, whereupon the r.b.c. uptake fell to 13.3 per cent, and once again improvement ensued and has been maintained for two months, to date.

DISCUSSION

Our data demonstrate 2 abnormalities of thyroid metabolism in 39 euthyroid patients with atrial fibrillation or tachycardia. The association of hyperthyroidism and atrial fibrillation has been observed so frequently, particularly in patients over 45 years of age (2, 12), that the presence of fibrillation constitutes a recognized indication to search for evidence of thyroid overactivity. Occasionally masked hyperthyroidism has been re-

vealed in this manner (13). All the patients in this study were therefore carefully evaluated and, utilizing the usual parameters, were described as euthyroid.

The rate of turnover of infused I^{131}-thyroxine was significantly increased over the mean for euthyroid subjects described in all published series (9, 10, 14), but it was not so rapid as has been observed in hyperthyroidism. Increased rates of turnover of infused thyroxine have also been observed in non-endocrine hypermetabolic states such as renal infection and leukemia (14). Except for the apprehensive subject mentioned previously, no patient in our series had an elevated BMR. Data consistent with increased disappearance rates of thyroxine have also been noted in animals after stress (15). These patients were under the stress of investigation, and the added discomfort of atrial fibrillation was present in some of them during the period of observations. Further studies are necessary in interim periods to clarify whether these phenomena are adequate to explain the increased turnover.

Altered rates of disposition of another labeled substance, I^{131}-albumin, have been found in congestive heart failure (16). Of the 11 infusion studies reported here, none was conducted in a patient with congestive failure. Four patients with paroxysmal atrial fibrillation who suffered from congestive failure were not included. The turnover of radiothyroxine was delayed in 3 of these 4 patients; in the fourth (a case of unexplained clotting defect), the half-time of disappearance of I^{131}-thyroxine was 5.1 days.

The apparent volume of the thyroxine distribution space was also increased. Consequently, despite the normal level of serum PBI, the product of the daily turnover of thyroxine, the circulating PBI, and the volume of the thyroxine distribution space, *i.e.*, the daily degradation of thyroxine, was also increased significantly above normal values. This degradation rate did not reach the levels found in thyrotoxicosis, but in 3 cases, it was greater than 100 micrograms daily.

The mean *in vitro* r.b.c. uptake of I^{131}-triiodothyronine, regarded as a measure of the relative binding affinities for thyroid hormone of plasma proteins and tissue components, was significantly elevated in the entire group, and in 13 patients it was in the range customarily associated with thyrotoxicosis (9). None of these patients was thyrotoxic. Similarly, high values were also found in patients with abnormalities of plasma proteins, those with carcinomatosis, severe liver or kidney disease and in some receiving anticoagulant theory (11). None of these conditions was present in our series of patients. Studies are in progress to learn whether changes in the thyroxine-binding moieties of plasma, such as have been described for thyrotoxicosis, are present in these subjects.

The relation of the r.b.c. uptake to the tachycardia is particularly strik-

ing in the patients treated with iodides. When iodide was administered in large doses, the r.b.c. uptake was decreased, and concomitantly there was a striking decrease in the paroxysms of atrial fibrillation. There are, as yet, inadequate data to indicate whether spontaneous cessation of paroxysms is associated with decreased red blood cell uptake. The mechanism of the action of iodides under these circumstances is unknown. Iodide is known to have the following effects on thyroid function: 1) block the synthesis of thyroxine, 2) reduce hormonal secretion from the thyroid 3) inhibit the action of thyrotropin, and 4) inactivate thyrotropin. It does not seem likely that these effects are pertinent here. On the other hand, little is known of the effect of iodide on the peripheral metabolism of thyroid hormone, on its protein binding, or on the permeability of cells thereto. Further studies are necessary to clarify this action of iodide.

Both turnover rates and changes of the relative plasma-tissue affinities for hormones indicated by the red blood cell data suggest increased availability of thyroid hormones to tissues but fail to explain why this increased availability did not result in general hyperthyroidism but affected only the heart. It is tempting to postulate a selective sensitivity of the heart to thyroid hormones. However, it is likely that local factors altering myocardial metabolism play a role in addition to neural and hormonal influences. It is thus significant that the patients with paroxysmal atrial fibrillation were in an age group (40 to 80 years) in which an underlying substrate of coronary atherosclerosis is likely.

Animal studies relating the rate of cardiac metabolism to administration of thyroid have yielded conflicting data, perhaps because of variation in experimental design. Gordon and Heming (17) found that thyroxine administered to rats in large doses for two or three days increased the oxygen consumption of excised, sliced or minced liver, kidney and diaphragm, more than that of the heart. McEachern (18) administered thyroxine over a longer period and found that the auricles from thyrotoxic animals consumed more oxygen than those from normal animals. Following single injections of thyroxine into thyroidectomized rats, Barker and Klitgaard (19) obtained a quicker and larger proportional increase in oxygen utilization by slices of left ventricular muscle than by any other tissue. Perhaps more relevant to the genesis of atrial tachycardia, Ulrick and Whitehorn (20) using rats made hyperthyroid by feeding desiccated thyroid for sixteen days found respiration increased 77 per cent in the auricles compared to 22 per cent in ventricular and 33 per cent in diaphragmatic muscle, whereas in skeletal muscle and small intestine there was no increase. Alternatively, thyroxine may potentiate the response to adrenaline (21). Moreover, this potentiation can be observed even before the calorigenic effect of thyroxine is evident (22, 23). In particular, thyroxine has been shown

to potentiate the increase in rate and metabolism of terrapin hearts induced by adrenaline (24). Whether thyroxine, in addition, potentiates the cardiac response to vagal substances such as acetylcholine and acetyl-β-methylcholine is unclear. Surtshin and Rucknagel (25) were unable to show that hyperthyroidism, induced in mongrel dogs by feeding or injecting thyroxine, caused increased sensitivity to acetyl choline. But Nahum and Hoff observed increased vagal effects in hyperthyroid patients, and in 4 instances were able to produce brief episodes of atrial fibrillation following injection of acetyl-β-methylcholine (26). Prolongation of the electrocardiographic PR interval, also a manifestation of increased vagal activity, has been demonstrated in thyrotoxicosis (27).

Clinical experience suggests other circumstances in which the particular sensitivity of the heart to thyroxine is evident. When large doses of desiccated thyroid are administered to euthyroid patients, palpitation may be a manifestation of toxicity before elevation of the basal metabolic rate occurs (28). Recent studies with the acetic-acid analogue of triiodothyronine have shown that it may induce angina pectoris in euthyroid and hypothyroid patients with coronary artery disease before there is any elevation of the general body metabolism (29). Moreover, when hypothyroidism is induced with I^{131} for the relief of intractable angina pectoris, close observation of the patient will occasionally reveal relief of pain shortly before the fall of the metabolic rate (30).

Thus, a variety of clinical and experimental data indicate an important role for thyroxine in the control of cardiac action and suggest possible mechanisms by which even small changes in the amount of thyroid hormone available to the tissues may increase cardiac metabolism and irritability and produce fibrillation or tachycardia without inducing the classic manifestations of hyperthyroidism.

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THE PLASMA PROTEIN-THYROID HORMONE COMPLEX IN MAN. III. FURTHER STUDIES ON THE USE OF THE *IN VITRO* RED BLOOD CELL UPTAKE OF I^{131}-*l*-TRIIODOTHYRONINE AS A DIAGNOSTIC TEST OF THYROID FUNCTION*†

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ABSTRACT

An analysis is presented of over 2900 determinations of the *in vitro* red blood cell uptake of I^{131}-*l*-triiodothyronine from whole blood and its correlation with thyroid status in man. The results indicate that this simple and rapid test, which avoids the administration of radioactivity to the patient, possesses a diagnostic accuracy comparable to other standard methods. It permits the assessment of thyroid function under circumstances in which other standard methods may not be applicable (particularly after the administration of organic iodine-containing compounds). It is useful in following the course of therapy for hyper- and hypothyroidism. The uptake is decreased in normal pregnancy and after administration of estrogen; lack of a decrease during pregnancy may presage miscarriage. The uptake is increased in nephrosis, in certain cases of liver disease and metastatic malignancy, in pulmonary insufficiency with CO_2 retention, in paroxysmal atrial arrhythmias, and following administration of anticoagulants. It is believed that this method is a practical and useful additional tool for the diagnosis, therapy, and further investigation of thyroid physiology and disease.

IN A previous publication (1), we reported our early results on the use of red blood cell *in vitro* incorporation of I^{131}-*l*-triiodothyronine from whole blood as an index of the status of thyroid function in man. This

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report is based on an analysis of over 2900 observations obtained over a four-year period in the routine clinical application of this method as another parameter of thyroid function. The results indicate that this red blood cell uptake test (a) is simple and rapid, (b) yields results with a diagnostic accuracy comparable to that of other standard methods, (c) permits assessment of thyroid function under conditions which preclude the application of other standard methods, (d) is useful in following the therapeutic course of hyperthyroidism and hypothyroidism, and (e) may serve as another meaningful parameter in the study of thyroid physiology and disease.

MATERIALS AND METHODS

Donor subjects consisted of healthy technicians, physicians and nurses, and patients from the Medical, Surgical, and Obstetrical Services, the Endocrine, Prenatal, and Radioisotope Clinics of the Beth Israel Hospital, children from the Children's Medical Center, and patients from various Boston hospitals. Studies were also carried out on blood samples mailed from distant areas. Thyroid status was determined, independently of the *in vitro* studies, by clinical evaluation (including subsequent response to therapy) and one or more standard laboratory procedures, *viz*, basal metabolic rate, serum protein-bound iodine level (PBI), serum butanol-extractable iodine level (BEI), 24-hour thyroidal uptake of radioactive iodine (I^{131}), salivary I^{131} excretion, plasma I^{131} conversion ratio, and blood cholesterol concentration.

The basic *in vitro* method was that previously described (1), namely (a) the addition of tracer amounts ($10–120 \times 10^{-4}$ μg.) of I^{131}-triiodothyronine (I^{131} TRI)[1] to 3-ml. aliquots of whole blood in a stoppered, 10-ml. Erlenmeyer flask, (b) incubation, with shaking, at 37° C. for two hours, (c) determination in a well-type scintillation counter of the radioactive content of two 1-ml. aliquots of whole blood (at counting rates sufficient to permit ± 2 per cent accuracy), (d) determination of the radioactive content of the red blood cell (r.b.c.) mass in each 1-ml. aliquot of whole blood aliquot after five washings in tenfold dilutions of isotonic sodium chloride, (e) calculation by the formula,

$$\% \text{ uptake} = \frac{\text{net counts in r.b.c.} \times 100}{\text{net counts/ml. of whole blood}},$$

and (f) correction of the percentage uptake to 100 hematocrit for comparative purposes.

RESULTS

Significant incorporation of I^{131} TRI by erythrocytes (5–8 per cent per 100 hematocrit), persisting after several washings, was observed as early as ten minutes, the shortest interval studied. Diagnostically differential results were obtained within fifteen to thirty minutes of incubation (Table 1). In exploratory studies, based on the foregoing basic method, reproducible and comparable results were obtained following addition of I^{131} TRI to as little as 0.5-ml. aliquots of blood in a stoppered small test tube (13 mm. × 100 mm.) used for subsequent incubation, counting and washing, thus

[1] Obtained from Abbott Laboratories, Chicago, Illinois.

TABLE 1. THE *in vitro* RED BLOOD CELL UPTAKE OF I^{131}-*l*-TRIIODOTHYRONINE (TRI) AT VARYING PERIODS OF INCUBATION OF THIS COMPOUND IN WHOLE BLOOD FROM EUTHYROID AND HYPERTHYROID SUBJECTS

Incubation period (hrs.)	Uptake of I^{131} TRI (%/100 hematocrit)	
	Euthyroid	Hyperthyroid
$\frac{1}{4}$	10.3	17.4
$\frac{1}{2}$	11.8	20.2
1	12.6	21.9
2	13.3	23.9
3	15.4	26.8
4	15.5	28.0
5	16.8	28.8

obviating the pipetting and transfer after incubation involved in the standard method. Reproducible results were obtained with routine, non-sterile bloods stored in the refrigerator for seven days and with sterile bloods stored under refrigeration for as long as thirty-two days. Results consistent with the reported thyroid status were obtained in samples mailed without any specific precautions from distant areas. Consistent results after correction to 100 hematocrit were obtained with bloods over a hematocrit range from 30 to 55. Variable results were sometimes found using bloods with hematocrit values outside of this range; such bloods were, therefore, corrected before final determination of uptake to hematocrit values within this range by the removal or addition, respectively, of appropriate aliquots of the donor's plasma prior to the addition of I^{131} TRI and incubation. Inconsistent results were obtained on a few occasions in bloods with a very rapid sedimentation rate; this was obviated by careful attention to provide adequate mixing immediately prior to each pipetting step.

In a series of 300 consecutive uptakes, the differences between duplicate determinations ranged from 0.0 to 1.6 per cent, averaging 0.6 per cent with a standard deviation[2] of ± 0.3. In 6 series involving 8 to 16 determinations each, the maximum standard error of the mean[3] was 3.8.

Effect of thyroid status

The results are summarized for females in Table 2 and Figure 1, and for

[2] Standard deviation $= \sqrt{\frac{\Sigma d^2}{n-1}}$

[3] Standard error of the mean $= \frac{\text{Standard deviation}}{\sqrt{\text{No. of observations}}}$

TABLE 2. SUMMARY OF THE TWO-HOUR *in vitro* RED BLOOD CELL UPTAKE OF I^{131} TRI FROM WHOLE BLOOD IN FEMALE SUBJECTS

Thyroid status	No. of observations	Uptake (%/100 hematocrit)
Euthyroid	1301	11.0–17.0 (Avge. = 13.9)
	3 (.2%)	<11.0
	49 (3.6%)	>17.0
Hyperthyroid	211	17.0–35.0 (Avge. = 22.5)
	2 (.9%)	<17.0
Hypothyroid	129	6.1–11.0 (Avge. = 9.3)
	1 (.7%)	11.1

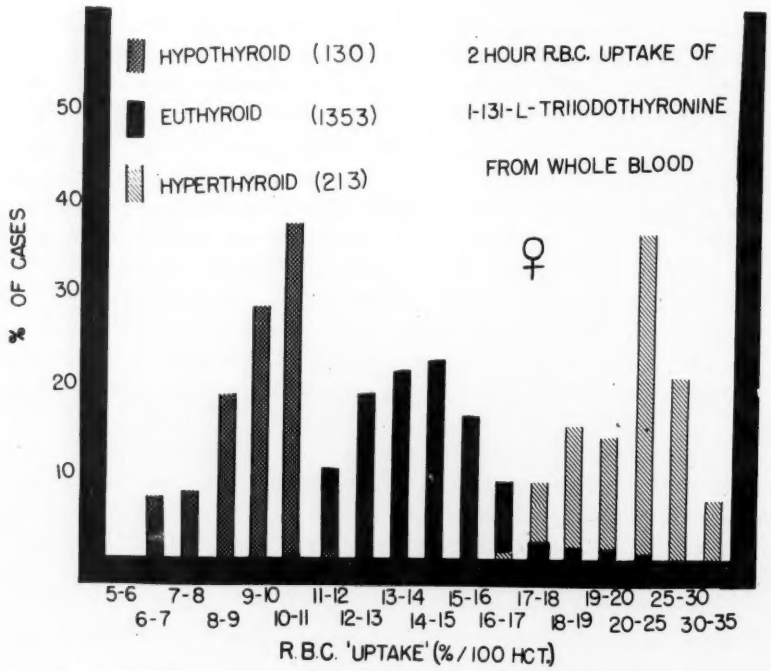

FIG. 1. Bar-graph summary of two-hour *in vitro* red blood cell uptake of I^{131} TRI in females. The height of each bar represents the percentage of patients in that group (hypothyroid, euthyroid or hyperthyroid) with uptakes within the range indicated at the base of the bar.

TABLE 3. SUMMARY OF THE TWO-HOUR *in vitro* RED BLOOD CELL UPTAKE OF I^{131} TRI FROM WHOLE BLOOD IN MALE SUBJECTS

Thyroid status	No. of observations	Uptake (%/100 hematocrit)
Euthyroid	566	11.8–19.0 (Avge. = 15.2)
	24 (4.1%)	>19.0
Hyperthyroid	64	19.5–37.9 (Avge. = 23.7)
	2 (3.0%)	<19.5
Hypothyroid	65	5.5–11.6 (Avge. = 9.7)

males in Table 3 and Figure 2. All uptake results are to be read as percentage uptake per 100 hematocrit for a two-hour incubation period.

Euthyroid females. In 1301 of 1353 determinations, the two-hour r.b.c. I^{131} TRI uptakes ranged from 11.0 to 17.0 per cent, averaging 13.9 per cent. The uptake was less than 11.0 per cent in 3 patients (0.2 per cent of the group) and greater than 17.0 per cent in 49 patients (3.6 per cent).

Hyperthyroid females. In 211 determinations, uptakes ranged from 17.0 to 35.0 per cent, averaging 22.5 per cent. In 2 subjects (0.9 per cent of the group), the uptake was less than 17.0 per cent. Definitive follow-up studies were possible in only 1 of these subjects, and it is of interest that her uptake, in the attained euthyroid state, was 9.9 per cent—distinctly below the range of normal.

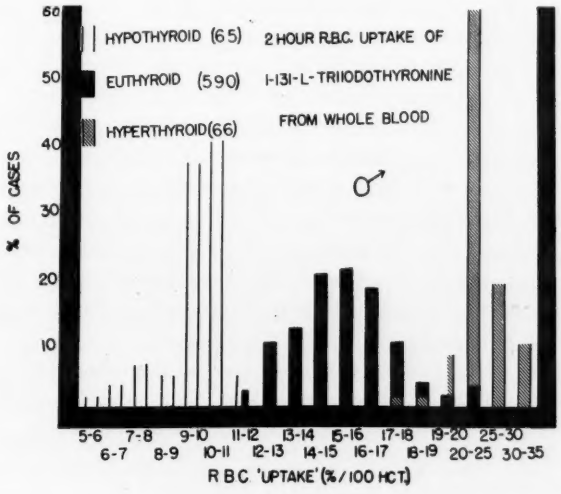

FIG. 2. Same as Figure 1, for male subjects.

Hypothyroid females. In 129 determinations, uptakes ranged from 6.1 to 11.0 per cent. In 1 subject, the value was 11.1 per cent.

Euthyroid males. In 566 determinations, uptakes ranged from 11.8 to 19.0 per cent, averaging 15.2 per cent. In 24 patients (4.1 per cent of the group), the uptakes were greater than 19.0 per cent.

Hyperthyroid males. In 64 determinations, uptakes ranged from 19.5 to 37.9 per cent, with an average of 23.7 per cent. In 2 patients (3.0 per cent of the group), the uptakes were less than 19.5 per cent.

Hypothyroid males. In 65 determinations, all uptakes ranged from 5.5 to 11.6 per cent, averaging 9.7 per cent.

The statistical validity of these suggestively higher values in males has not yet been determined. Thus, of 1943 euthyroid subjects, 76 (3.9 per cent) had r.b.c. I^{131} TRI uptake values which fell outside the selected euthyroid limits. These 76 cases are considered further under the subsequent analysis of certain of the factors affecting r.b.c. uptake. The remaining 1867 cases included subjects with no known abnormality, patients with nontoxic nodular goiter, colloid goiter, thyroid carcinoma, and various nonthyroid conditions such as diabetes mellitus, Addison's disease, hypertension, congestive heart failure (including cases treated with mercurials), severe anxiety states, polycythemia vera, hepatolenticular degeneration (Wilson's disease), or severe malnutrition.

Of 279 hyperthyroid patients, 4 (1.4 per cent) had r.b.c. uptakes which were below the upper euthyroid limit.

Of 195 hypothyroid patients, only 1 (0.5 per cent) had an r.b.c. uptake above the lower euthyroid level. Diagnostically low values were obtained in all cases of spontaneous myxedema, panhypopituitarism and cretinism; and ultimately they were obtained in hypothyroidism which developed following therapy of hyperthyroidism by surgery, propylthiouracil or radioactive iodine. In several euthyroid patients in whom myxedema was purposely being induced by administration of I^{131} (repeated doses of 10–20 millicuries) for therapy of severe angina pectoris or congestive heart failure, there was a discrepant time-lag; r.b.c. uptakes remained in the euthyroid range at a time when the patients exhibited suggestive or definite clinical and (or) laboratory evidence of the hypothyroid state. On continued observation, if therapy was withheld or inadequate, the r.b.c. uptakes ultimately (six to eighteen months) fell to low levels.

The results of other laboratory tests were diagnostically distinctive, in varying degrees, in the 3 major categories of thyroid function. However, there was no apparent correlation within any single category between the absolute r.b.c. I^{131} TRI uptake value for given patients and their BMR, serum PBI level, or 24-hour thyroidal I^{131} uptake.

Effect of therapy for hyperthyroidism

Repeated observations were carried out on 76 hyperthyroid patients, during and after therapy with stable iodides, propylthiouracil, surgery or radioactive iodine. In 4 instances, the r.b.c. uptake of I^{131} TRI persisted in the hyperthyroid range for as long as one year after attainment of the euthyroid (3 patients) and hypothyroid (1 patient) states; subsequently (eighteen to twenty-four months after therapy) the r.b.c. uptakes were consistent with the attained clinical state in 3 of these subjects; the fourth was lost to follow-up. In all other instances, correlation of r.b.c. uptake and thyroid status was excellent. The administration of propylthiouracil often caused a decreased r.b.c. uptake as early as twenty-four to forty-eight hours after institution of therapy, before there was any apparent clinical improvement. Failure of r.b.c. uptake to decrease within one week presaged failure of clinical response, necessitating increased dosage. Similarly, failure of an increased r.b.c. uptake to reach normal levels within two to three months following surgery or administration of I^{131} was correlated in every instance (except for the 4 cases just mentioned) with the ultimate diagnosis of persistent hyperthyroidism. Decrease of uptake to hypothyroid levels was an early and reliable index of overtreatment. The *in vitro* addition to hyperthyroid bloods of large amounts of stable iodide or propylthiouracil failed to lower the subsequent r.b.c uptake. As in the untreated states, there was no apparent direct correlation between r.b.c. uptakes of I^{131} TRI and other parameters of thyroid function (BMR or serum PBI level). Although results obtained by these methods were usually in general agreement, in several instances the r.b.c uptake reached a level consistent with the evaluated thyroid status in advance of the other parameters.

Effect of therapy for hypothyroidism

Repeated observations were carried out during treatment of 41 adult hypothyroid patients with desiccated thyroid, *l*-thyroxine, *l*-triiodothyronine, or various combinations thereof. The over-all correlation was excellent. In every instance, ultimately adequate therapy resulted in a progressive increase of r.b.c. I^{131} TRI uptake to the normal range. Failure of attainment of normal levels reliably indicated undertreatment. Increase of values to levels greater than normal reliably indicated overtreatment. The method was of some distinctive value when therapy was undertaken with *l*-triiodothyronine, as in this instance serum PBI values are not diagnostically determinative.

With the cooperation of Dr. John Crigler of the Children's Medical Center, Boston, a long-term study was instituted of the treatment of cre-

tinism with the various thyroid hormones. R.b.c. uptakes of I^{131} TRI were measured repeatedly in more than 40 cretins over periods up to two years. From preliminary analysis of the data it appears that the r.b.c. uptake test is a very satisfactory method for following the therapeutic course. With adequate therapy, the r.b.c. uptake generally increased to the normal range. Again, the r.b.c. uptake was uniquely useful in following therapy in those cases in which *l*-triiodothyronine had been selected as the therapeutic agent; in these patients, the serum PBI level cannot be used and the BMR determination is not feasible. The studies are continuing and will be reported later.

Effect of exogenous iodine

Determinations were made in 107 cases in which evaluation was complicated by the known or assumed use of some form of organic iodine. In 42 subjects, studies were carried out before and after (four hours to two months) the administration of organic iodine-containing compounds (such as Priodax, Cholegrafin or Urokon), when the serum PBI levels and thyroidal I^{131} uptakes were spuriously abnormal. As previously reported (1), slight increases in the r.b.c. uptake of I^{131} TRI were observed in 4 subjects; results were unaltered in the other 38 subjects. In the remaining 65 of the 107 patients, control observations were not available. The r.b.c. uptake determinations were carried out after known or suspected iodine exposure at a time when the serum PBI levels were distinctly elevated (up to >100 μg. per 100 ml.) and the 24-hour thyroidal I^{131} uptake was suppressed below 10 per cent. Three of these subjects were clinically hyperthyroid, and the r.b.c. uptake was in the hyperthyroid range (22.0 to 25.2 per cent). Among the remaining 62 subjects, all thought to be euthyroid, r.b.c. uptakes were normal in 60 and elevated in 2 (22.4 and 24.2 per cent). No known originally hypothyroid subjects given exogenous iodine were available for study.

The administration of iodides to hyperthyroid subjects caused a progressive decrease in the r.b.c. uptake of I^{131} TRI. When given to euthyroid subjects, iodides caused either no change or, after very large doses, a distinct decrease.

The case of Mr. B.F., aged 55, is of particular interest. He presented with the classic picture of myxedema. The r.b.c. uptake of I^{131} TRI, however, was 12.7 per cent (a normal value). His BMR was -22 per cent, blood cholesterol level 384 mg. per 100 ml., and 24-hour thyroidal I^{131} uptake 5.2 per cent. The serum PBI level was reported as "way over" 20 μg. per 100 ml. Further questioning revealed that he had been ingesting 3 to 4 teaspoonsfuls of a cough syrup containing saturated solutions of potassium iodide daily for about seven years. On the assumption that

this represented a case of myxedema due to excess iodide administration, he was instructed to omit all medication. There was a prompt and progressive return to the euthyroid state. Laboratory studies two months later revealed a BMR of −1 per cent, a blood cholesterol level of 218 mg. per 100 ml., a serum PBI level of 6.6 μg. per 100 ml., and a 24-hour thyroidal I^{131} uptake of 28 per cent. Throughout this entire course, the r.b.c. uptake remained between 12.0 and 16.9 per cent on 6 occasions. One year later, he was still completely euthyroid; his r.b.c. uptake of I^{131} TRI was 16.8 per cent.

Effects of pregnancy and of estrogen

As previously reported (1), the r.b.c. uptake of I^{131} TRI was consistently decreased during normal pregnancy (142 determinations during 48 pregnancies). The time of onset of this decrease has not been precisely defined. In 1 patient there was a decrease from 15.3 per cent before pregnancy to 8.5 per cent at the estimated third week of gestation, and in 1 patient there was a decrease from 12.2 to 9.1 per cent between the fourth and seventh weeks. In several instances values were decreased between the sixth and seventh week, and from this time on, all uptakes ranged between 4.7 and 11.8 per cent, averaging 7.4 per cent. Uptakes in each subject remained about the same throughout pregnancy; there was no significant difference between the average values for each trimester. Following delivery, the uptake returned to normal between the first and second week post partum. In 2 patients, there was an apparent increase to hyperthyroid levels on the fifteenth and seventeenth days, respectively. In 6 pregnancies, the r.b.c. uptake was found to be in the normal, euthyroid range beyond the eighth week of gestation. Each of these pregnancies subsequently terminated in miscarriage. In 1 patient who had had repeated miscarriages in previous pregnancies, premature delivery of a nonviable fetus occurred during the seventh month, despite the usual decreased r.b.c. uptake after the second month.

On repeated observations, r.b.c. uptake was decreased following the administration of estrogens to male or female subjects, but not following cortisone or chorionic gonadotropin.

Effect of anticoagulants

The previously observed increase in r.b.c. uptake of I^{131} TRI after administration of Dicumarol (1) has been confirmed but was not observed as consistently as in the prior small series. Studies were carried out in 66 subjects. In 53, the r.b.c. uptake was increased. Elevated values were observed as early as twenty-four to forty-eight hours after administration of 200–300 mg. of Dicumarol. Values increased from control levels of 11.0–16.4 per

cent to 17.8–31.9 per cent. Once increased, the levels remained essentially the same through the period of treatment. Adequate follow-up data after omission of therapy was not obtained but, in a small series, uptakes had returned to or towards normal by the end of fourteen to eighteen days.

Comparable increased r.b.c. uptakes were observed in 8 patients treated with heparin, in 2 treated with Tromexan, and in 2 treated with phenylindandione. There was no apparent correlation of r.b.c. uptake levels with prothrombin concentration. Uptakes were normal in 2 euthyroid subjects with deficiency of plasma thromboplastin component and in 1 with deficiency of plasma thromboplastin antecedent.

Effects of miscellaneous factors

The r.b.c. uptake of I^{131} TRI was consistently increased in nephrosis, and frequently increased in severe hepatic disease (fulminant hepatitis, marked cirrhosis) and extensive metastatic malignancy.

Another common denominator in the group of euthyroid subjects with increased uptakes is represented by 8 patients with severe, long-standing pulmonary insufficiency with marked CO_2 retention. Uptakes in this group ranged from 22.4 to 37.1 per cent. Although the reason for this effect is not clear, it may be pertinent to consider here certain experimental studies on the effects of altered CO_2 concentration on the r.b.c. uptake of I^{131} TRI. These studies were first suggested by an observation of Dr. A. Ureles and Dr. M. Murray, Rochester, N.Y. In a personal communication relating to their r.b.c. uptake studies, they noted that the uptake was strikingly decreased when the incubation was carried out in unstoppered flasks. We have confirmed their observation. In a series of 32 experiments in parallel, the r.b.c. uptake determined by incubating the blood in open flasks was only 40–50 per cent of that determined on the same blood incubated in stoppered flasks. This effect was observed in all types of blood studied, whether from euthyroid, hyperthyroid, hypothyroid or pregnant patients. When 5 per cent CO_2 was bubbled into the *open* flask during incubation, this effect was overcome and values identical with those determined in the stoppered flask were obtained. In other experiments, unstoppering the flask after a two-hour incubation and continuing the run for two more hours led to the same striking decrease from the normal values attained in the duplicate flask, stoppered throughout. Stoppering the flask after an initial incubation in an open flask resulted in a progressive increase to normal control values compared with the decreased level persisting in the duplicate flask, unstoppered throughout. R.b.c. uptakes in euthyroid blood could be increased to the hyperthyroid range by bubbling 5 per cent CO_2 into the *stoppered* flask during the incubation period. Such increases, however, were relatively small (18.0 to 20.0 per cent) in comparison with the much greater increase in uptake observed in

the majority of the 8 patients with *in vivo* CO_2 retention. Studies are continuing on the role of CO_2 (pH, HCO_3) in the incorporation of labeled thyroid hormones into red blood cells.

A final categorization of increased r.b.c. uptake of I^{131} TRI in otherwise euthyroid subjects is represented by the investigation in our laboratories of euthyroid patients with paroxysmal atrial arrhythmias. A detailed report appears elsewhere in this issue (2). Of pertinence here is the finding that of 18 males studied, 8 had r.b.c. uptakes greater than 19.5 per cent; and of 21 females studied, 7 had r.b.c. uptakes greater than 18 per cent. In 12 of the 15 patients with high uptakes the diagnosis was paroxysmal atrial fibrillation; in 3 the diagnosis was paroxysmal atrial tachycardia.

DISCUSSION

Although there has been a significant advance in our understanding of certain of the parameters of thyroid function, the mechanism of incorporation of thyroid hormone by the "peripheral" tissues and its role in over-all thyroid economy remain to be elucidated. Growing importance has been attributed to the role of the plasma protein-thyroid hormone complex. Recent studies (3–12) employing radioactive, electrophoretic techniques have demonstrated that thyroxine, quantitatively the major circulating hormonal component, is bound primarily to a more or less specific protein moiety which migrates electrophoretically, at pH 8.6, between the alpha-1 and alpha-2 globulins (the thyroxine-binding globulin). Other, less extensive studies (11,12) indicate that triiodothyronine is similarly bound, although less completely and less firmly. Small amounts of thyroxine are bound to albumin which serves as the major secondary carrier, particularly when thyroxine levels are increased. It is assumed that, as more thyroxine is added, the binding sites of the inter-alpha globulin are more fully occupied and additional thyroxine then "spills over" onto albumin which, in turn, has a weaker binding affinity for thyroxine. Data concerning the sites or mechanism of cellular or tissue binding of thyroxine are not available. It has been suggested that the distribution of thyroid hormones between serum and cells is dependent upon the relative binding capacities for thyroid hormone of the serum protein(s) and some binding factor (? protein) in the cell (13–15).

According to this concept, the *in vitro* r.b.c. uptake may be determined, in part, by the availability (and affinity) of binding sites for the added I^{131} TRI. If more of the sites are occupied because of the increased thyroxine levels in hyperthyroidism, more of the added I^{131} TRI will spill over to the weaker secondary carrier(s) and thus more will be taken up by the cell. The opposite would obtain in myxedema, with more (or firmer) binding sites available. The decreased r.b.c. uptakes in pregnancy and the increased uptakes in nephrosis would be explained by the respective increase

(16–18) and decrease (19) of specific plasma binding globulins which have been demonstrated in these conditions. Further studies are necessary to assess the validity of this concept in the explanation of the herein reported results obtained during iodine or Dicumarol therapy and in such conditions as CO_2 retention, liver disease, or paroxysmal atrial arrhythmias.

More recent studies by others, and further observations in our laboratory, however, raise serious questions as to the validity of this attractive, simplified concept. The specific binding globulin has not yet been definitively isolated and characterized in order to permit its precise quantitation in the various states of thyroid function. The homogeneity, specificity, and actual role of the thyroid hormone-binding protein needs re-evaluation in the light of newer data resulting from more recent refinements in techniques. Robbins and Rall (20), using a reverse-flow electrophoresis technique and mathematical calculations based on several fundamental assumptions, assigned a major role to the level of "free" thyroxine in determining availability and tissue uptake of thyroid hormone. Ingbar (21), using a new electrophoretic system, found a major binding protein in the pre-albumin area in addition to the inter-alpha globulin. Rich and Bearn (22), using a starch-gel electrophoresis method after Smithies (23), observed that the specific thyroxine-binding protein is localized only to the first pre-albumin band. In our laboratory (24), combining starch-gel electrophoresis and tris(hydroxymethyl)-aminomethane buffer, we found 3 distinctive binding sites for thyroxine: 1) a pre-albumin area, 2) an albumin area (which on transfer to paper and subsequent electrophoresis migrates in a sharp band behind, not with, the major stainable albumin peak), and 3) an inter-alpha globulin area.

Such results and their rapid unfolding with technical developments necessitate caution in attempting to effect physiologic interpretations of complex *in vitro* interrelationships in terms of probably different, equally complex physico-chemical determinants of protein-protein interactions.

Despite such limitations of precise delineation of the mechanism of tissue incorporation of thyroid hormone, our continuing clinical experience indicates that the described r.b.c. *in vitro* uptake of I^{131} TRI is a meaningful additional tool for the diagnosis, therapy and investigation of thyroid physiology and disease. The method is simple and rapid, and it avoids the administration of radioactivity to the patient. Its diagnostic accuracy compares favorably with that of other currently used methods (25). We have found it very useful as a rapid screening method. It permits the assessment of thyroid function under certain circumstances where other standard methods may not be applicable (Table 4). In particular, it appears to be relatively unaffected by the prior administration to the patient of organic iodine-containing compounds. It is useful in following and gauging the adequacy of therapy of hyperthyroidism and hypothyroidism, particu-

TABLE 4. SUMMARY OF THE EFFECT OF VARIOUS FACTORS ON THE TWO-HOUR *in vitro* RED BLOOD CELL UPTAKE OF I^{131} TRI FROM WHOLE BLOOD

I. *Decrease*
- A. Pregnancy
- B. Estrogen
- C. Propylthiouracil in hyperthyroidism
- D. Iodide in hyperthyroidism

II. *No (or slight) effect*
- A. Thyroid enlargement—colloid goiter, nontoxic nodular goiter, thyroid carcinoma
- B. Exogenous iodine
- C. Anxiety
- D. Congestive heart failure
- E. Hypertension
- F. Administration of mercury
- G. Other endocrinopathies—diabetes mellitus, Addison's disease

III. *Increase*
- A. Anticoagulants—Dicumarol, heparin
- B. Nephrosis
- C. Severe liver disease
- D. Severe metastatic malignancy
- E. Severe pulmonary insufficiency with CO_2 retention
- F. Paroxysmal atrial arrhythmias

larly, in the latter instance, when *l*-triiodothyronine is used. Its alteration in certain cases of abnormal pregnancy, anticoagulant therapy, liver disease, metastatic malignancy, CO_2 retention, and paroxysmal atrial arrhythmia indicates areas for continuing investigation. The final value of this technique will be determined by the results of its use in various clinics —the ultimate test, which we believe is merited by the data presented.

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THE EFFECT OF *l*-THYROXINE ON SERUM LIPOPROTEIN AND CHOLESTEROL CONCENTRATIONS*

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ABSTRACT

The effect of intravenous administration of 6 mg. per week of *l*-thyroxine for a period of sixteen weeks on low-density and high-density serum lipoproteins, total serum cholesterol, body weight, pulse and blood pressure was studied in a group of 13 institutionalized ambulatory schizophrenic patients.

S_f°0–12 and S_f°12–20 serum lipoprotein concentrations were significantly reduced, but no effect on S_f°20–100 and S_f°100–400 was observed. Total serum cholesterol as well as high-density serum lipoprotein concentrations were also significantly reduced. The reduction in low-density serum lipoprotein concentrations was found to be highly correlated with initial (pre-treatment) levels, resulting in greater decreases in serum lipoprotein concentrations the higher the initial level.

Body weight and diastolic blood pressure decreased significantly and pulse rate increased.

l-Thyroxine is an effective hypolipoproteinemic substance, producing a pattern of changes in serum lipoprotein and total serum cholesterol similar to that previously reported with desiccated thyroid.

AMPLE evidence (1–7) is available to indicate that desiccated thyroid, *l*-thyroxine and *l*-triidothyronine have similar metabolic effects.

Our studies using desiccated thyroid at a dosage level of 325 mg. per day have demonstrated profound and maintained decreases in the concentrations of the S_f° 0–12 and S_f° 12–20 low-density serum lipoproteins and of serum cholesterol (8). These results led us to an investigation of the effect of *l*-thyroxine on serum lipoprotein and serum cholesterol concentrations.

MATERIAL AND METHODS

A group of schizophrenic patients at Napa State Hospital was chosen for this study. These patients were physically and metabolically normal insofar as could be determined by a physical examination and routine laboratory tests. Their psychiatric status

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made leave or discharge from the hospital unlikely. None of the patients had been receiving any medication prior to the administration of *l*-thyroxine.

The group consisted of 8 males and 8 females, of mean ages 35.5 and 38.9, years, respectively. All were between 26 and 49 years of age. A control group of 2 male and 2 female patients (ages 20, 38, 34 and 51 years, respectively), not receiving any medication, was also studied.

The 16 patients received weekly intravenous injections of 6 ml. of a solution of *l*-thyroxine (1 mg. per ml.) for sixteen weeks. A dose of 6 mg. per week of *l*-thyroxine was chosen because this amount was estimated to be equivalent to a dose of 325 mg. per day of desiccated thyroid (9). The 4 control patients received a weekly intravenous injection of 6 ml. of physiologic sodium chloride for sixteen weeks.

Three pre-therapy blood samples were drawn from each of the 20 patients at weekly intervals. During the 16-week period of *l*-thyroxine therapy, blood samples were drawn at approximate two-week intervals. The weekly injection of *l*-thyroxine was always administered *after* blood samples had been obtained. In the post-therapy period, blood samples were drawn two, four, and eighteen weeks after the cessation of *l*-thyroxine administration.

All blood samples were analyzed for concentration of low-density serum lipoproteins S_f° 0–12, S_f° 12–20, S_f° 20–100, and S_f° 100–400, for high-density lipoproteins HDL-1, HDL-2, and HDL-3, and for cholesterol. The analytical methods used have been described in detail elsewhere (10, 11).[1]

Weight, blood pressure and pulse were measured at the time of collection of each blood sample.

RESULTS

Of the original group of 16 patients, 3 did not receive the complete series of injections. The reasons for stopping the *l*-thyroxine were: technical difficulty in administration because of poor veins in 2 cases, and excessive weight loss in 1 case. The effects of *l*-thyroxine on serum lipoprotein concentration, blood pressure, pulse rate and weight in the 3 patients who received less than the total of 96 mg. were not different from those observed in the group of 13 patients who received the complete series of 16 injections.

In the discussion of the results which follows, an "effect" or "response" is defined as the change observed in the mean value for 7 measurements during *l*-thyroxine compared with the mean value for 3 pre-therapy measurements obtained on the same group of patients.

Low-density lipoproteins

The effect of *l*-thyroxine on the levels of low-density serum lipoproteins in the 13 patients (7 males and 6 females) who received the entire series of 16 injections are shown in Figure 1. The low-density lipoprotein levels in the 4 control patients who received injections of physiologic sodium chloride are also shown.

[1] The high-density lipoprotein analyses were made by Oliver deLalla, using the analytical method developed by him (10).

The concentrations of S_f° 0–12 and S_f° 12–20 serum lipoproteins immediately decreased to a highly significant degree (Table 1). The number of patients who showed decreases in lipoprotein concentrations are shown in Table 2. These marked decreases were maintained for the entire period of *l*-thyroxine administration. After cessation of *l*-thyroxine therapy the level of S_f° 0–12 rose immediately. After four weeks without *l*-thyroxine, the concentration of S_f° 0–12 had risen to slightly above the initial level, but

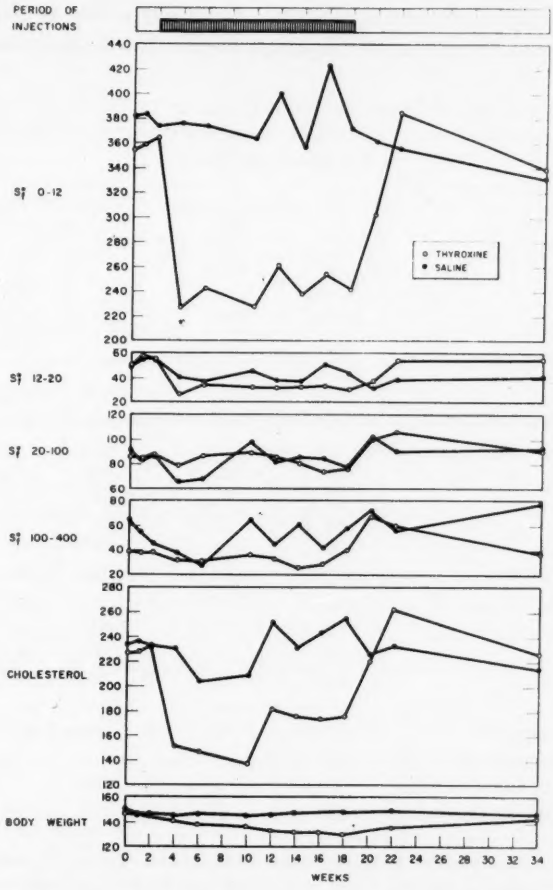

FIG. 1. Effect of *l*-thyroxine on the levels of serum low-density lipoprotein (S_f° 0–12, S_f° 12–20, S_f° 20–100, and S_f° 100–400) and cholesterol, and on body weight in a group of 13 patients. Four control patients received injections of physiologic sodium chloride. Serum lipoprotein and cholesterol concentrations are expressed as mg./100 ml., and body weight in pounds.

after eighteen weeks without *l*-thyroxine it had fallen to slightly below the initial value. The S_f° 12–20 lipoprotein concentration did not show a rebound above initial levels; it returned to the initial value after administration of *l*-thyroxine had been stopped for four weeks.

There were no significant changes in the S_f° 20–100 and S_f° 100–400 lipoprotein concentrations during *l*-thyroxine therapy, for the group as a whole. However, pronounced decreases did occur in some of the patients, particularly those who had high initial levels. Both the S_f° 20–100 and S_f° 100–400 lipoproteins showed a rebound above initial levels after two and four weeks without *l*-thyroxine. Eighteen weeks after *l*-thyroxine had been

TABLE 1. RESPONSES TO INJECTION OF *l*-THYROXINE IN 13 PATIENTS COMPARED WITH THE RESPONSES TO INJECTION OF PHYSIOLOGIC SODIUM CHLORIDE IN 4 CONTROL SUBJECTS

	l-Thyroxine			Control	
	(a)* Mean initial conc. (mg./100 ml.)	(b) Mean change (mg./100 ml.)	Signif. of change	(c) Mean initial conc. (mg./100 ml.)	(d) Mean change (mg./100 ml.)
Serum					
S_f° 0– 12	360.7	−121.0	P < .001	379.7	− 2.5
S_f° 12– 20	55.8	− 24.1	P < .01	54.1	−12.0
S_f° 20–100	86.7	− 5.3	ns†	87.9	− 8.5
S_f° 100–400	37.9	− 5.5	ns	54.8	− 7.4
S_f° 0–400	541.1	−155.8	P < .001	576.4	−30.3
HDL-1	23.2	− 4.2	P < .01	26.0	0.0
HDL-2	62.7	− 16.7	P < .01	48.0	− 1.0
HDL-3	218.1	− 35.6	P < .001	231.8	− 3.0
Cholesterol	229.1	− 65.5	P < .001	233.5	− 1.0
Body weight (pounds)	146.2	− 13.2	P < .001	147.8	− 1.6
Systolic blood pressure (mm.Hg)	125.4	− 0.2	ns	129.3	− 5.5
Diastolic blood pressure (mm.Hg)	78.3	− 10.8	P < .001	82.8	− 5.8
Pulse rate (beats/min.)	80.5	+ 16.7	P < .001	90.2	− 1.4

* (a) Initial values are the mean of 3 measurements on each of 13 patients.
(b) Mean change equals the mean of 3 initial values for 13 patients minus the mean for the 7 samples during thyroxine therapy for the same 13 patients.
(c) Initial values are the mean of 3 measurements on each of 4 patients.
(d) Mean change equals the mean of 3 initial values for 4 patients minus the mean for 7 control samples for the same 4 patients.

† Not significant.

TABLE 2. EFFECT OF *l*-THYROXINE ON VARIOUS MEASURES IN TERMS OF THE NUMBER OF PATIENTS SHOWING A RESPONSE

	Total No. of patients	No. of patients showing decreases	No. of patients showing increases
Serum			
S_f° 0– 12	13	13	0
S_f° 12– 20	13	11	2
S_f° 20–100	13	8	5
S_f° 100–400	13	8	5
S_f° 0–400	13	13	0
HDL-1	13	10	3
HDL-2	13	12	1
HDL-3	13	13	0
Cholesterol	13	13	0
Body weight	13	13	0
Systolic blood pressure	13	4	9
Diastolic blood pressure	13	13	0
Pulse rate	13	0	13

stopped, the concentrations of both had returned to the initial values.

Considering the entire low-density lipoprotein spectrum from S° 0 to $S_\circ$ 400, a marked decrease in concentration (28 per cent of the initial value) was observed. This change was significant at the 0.1 per cent level.

These responses to *l*-thyroxine were consistent with, and were of approximately the same magnitude as those observed with 325 mg. per day of desiccated thyroid, as previously reported by us (8).

The high correlation between the initial levels of all the low-density lipoproteins and extent of response to *l*-thyroxine deserves emphasis. As shown in Table 3, the correlation coefficients between initial level and response to *l*-thyroxine are higher for the S_f° 0–12 and S_f° 12–20 lipoproteins than for the S_f° 20–100 and S_f° 100–400 lipoproteins.

TABLE 3. RELATIONSHIP BETWEEN INITIAL SERUM LIPOPROTEIN CONCENTRATION AND RESPONSE TO *l*-THYROXINE

Class of lipoprotein	Correlation coefficient between initial concentration and response to *l*-thyroxine
S_f° 0– 12	+0.87
S_f° 12– 20	+0.98
S_f° 20–100	+0.69
S_f° 100–400	+0.60
S_f° 0–400	+0.79

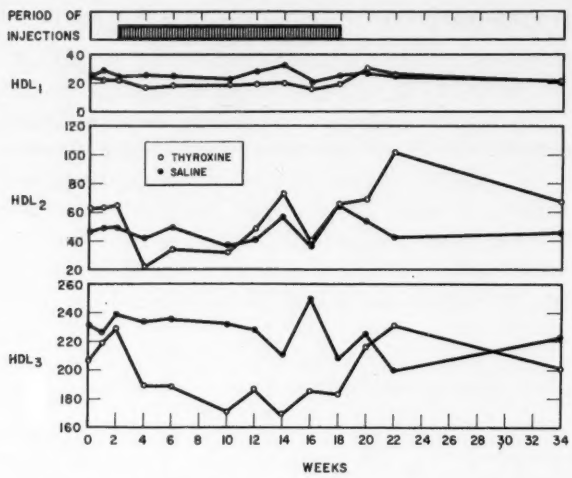

FIG. 2. Effect of *l*-thyroxine on the levels of serum high-density lipoproteins (HDL_1, HDL_2 and HDL_3) in a group of 13 patients. Four control patients received injections of physiologic sodium chloride. Lipoprotein concentrations are expressed as mg./100 ml. of serum.

These data show that patients with high initial concentrations of any or all of the low-density lipoproteins can be expected to respond with greater decreases than do patients with low initial concentrations.

Greater lipoprotein responses in patients with high initial levels were also observed in our previous studies on the effect of desiccated thyroid (8).

High-density lipoproteins

The 3 high-density lipoproteins, HDL-1, HDL-2 and HDL-3, decreased in concentration during the administration of *l*-thyroxine, as shown in Figure 2 and Table 1. The number of patients who showed such a decrease are shown in Table 2. High-density lipoproteins in the 4 control patients are also shown in Figure 2 and Table 1. The changes during thyroxine therapy were highly significant, P being less than 0.01 for HDL-1 and HDL-2, and less than 0.001 for HDL-3. The HDL-1, HDL-2 and HDL-3 lipoprotein levels rebounded above the initial concentrations in the period after omission of *l*-thyroxine; the rebound for HDL-1 was greatest at two weeks, and for HDL-2 and HDL-3 at four weeks after discontinuation. By the end of the period of study, after eighteen weeks without *l*-thyroxine, the concentrations of the high-density lipoproteins were again approximately the same as the respective initial values.

The response of the HDL-2 lipoprotein level to administration of *l*-

thyroxine was not maintained for the entire 16-week period. By the tenth week of *l*-thyroxine therapy, the HDL-2 lipoprotein concentration had returned to the initial value.

Cholesterol

Serum cholesterol concentrations decreased to a highly significant degree (P less than 0.001) while the 13 patients were receiving *l*-thyroxine. A rebound above initial concentrations was observed during the post-therapy period. After eighteen weeks without *l*-thyroxine, the cholesterol concentrations had returned to initial pre-therapy levels. These results are shown in Figure 1 and Table 1.

Body weight

During the period of *l*-thyroxine injections, body weight fell gradually from a mean initial value of 146.2 pounds. The mean weight loss was 13.2 pounds (Fig. 1 and Table 1). Eighteen weeks after omission of *l*-thyroxine the body weight had returned to 144.1 pounds, approximately the initial value.

Blood pressure and pulse

As shown in Table 1 and Figure 3, there was no change in systolic blood pressure as a result of administration of *l*-thyroxine.

Diastolic blood pressure decreased markedly during the period of *l*-thyroxine therapy; furthermore, this decrease was observed in *all* 13 patients. Menof (12) reported a lowering of diastolic blood pressure in re-

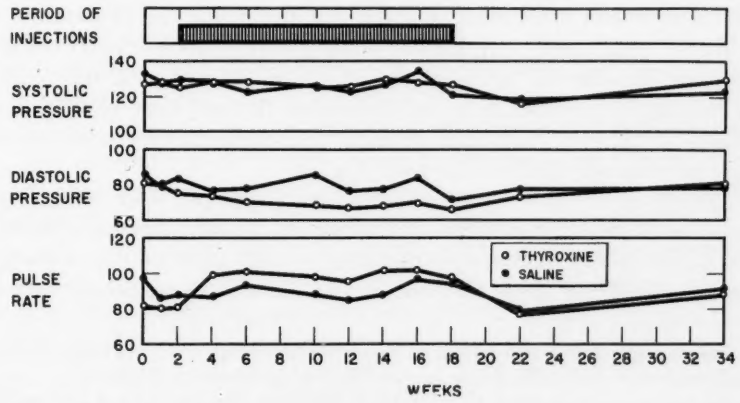

FIG. 3. Effect of *l*-thyroxine on blood pressure and pulse in a group of 13 patients. Four control patients received injections of physiologic sodium chloride. Blood pressure in terms of mm. Hg; pulse rate in beats/minute.

sponse to the administration of desiccated thyroid in doses of 130–390 mg. per day. However, using 325 mg. per day of desiccated thyroid, we were unable to demonstrate any change in diastolic blood pressure (8).

The pulse rate increased significantly in the 13 patients while they were receiving *l*-thyroxine. No definite relationship could be demonstrated between the decrease in diastolic blood pressure and the increase in pulse rate in this group of patients, even though all 13 patients had increases in pulse rate and decreases in diastolic blood pressure. There was a suggestive, but statistically non-significant relationship ($r=0.70$) between the decrease in S_f° 0–12 and S_f° 0–400 levels and the increase in pulse rate. The patients with the greatest decreases in the levels of these lipoproteins usually showed the smallest increases in pulse rate; and, conversely, those with minimal decreases in lipoprotein levels showed more marked increases in pulse rate than did the group as a whole.

DISCUSSION

The effect of *l*-thyroxine in a dosage of 6 mg. per week on the concentration of low-density serum lipoproteins was compared with the effect of various daily dosages of desiccated thyroid on these lipoproteins as follows:

The 13 patients receiving *l*-thyroxine were divided into 2 groups, one composed of the 7 patients with the highest initial levels of S_f° 0–400 lipoprotein and the other composed of the 6 patients with the lowest initial levels. The decreases in concentration were then plotted against the initial concentrations for the 2 groups.

Similarly, the 39 patients who received various dosages of desiccated thyroid were divided into 2 groups—the 20 patients with highest initial levels of S_f° 0–400 lipoprotein forming one group and the 19 patients with the lowest initial levels forming the other group. The decreases in S_f° 0–400 concentration effected by daily oral doses of 195 mg., 260 mg., 325 mg. and 390 mg. (approximately)[2] of desiccated thyroid were plotted against initial S_f° 0–400 concentrations. Straight lines were drawn connecting the 2 points at the same dosage of thyroid (Fig. 4).

Comparison of the curves indicates that 6 mg. per week of *l*-thyroxine administered intravenously has an effect on S_f° 0–400 lipoprotein concentrations which is approximately equivalent to that produced by a daily oral dose of 325–390 mg. of desiccated thyroid. Kirsch (9) previously estimated

[2] This is a calculated rather than an experimental dosage level, estimated as follows: Based on Greer's work (13), it was assumed that administration of 195 mg. of desiccated thyroid per day is equivalent to a total endogenously effective dose of 390 mg. for an initial period of a few weeks during which endogenous thyroid hormone production (calculated to be equivalent to about 195 mg. of desiccated thyroid per day) is not yet affected by administration of thyroid.

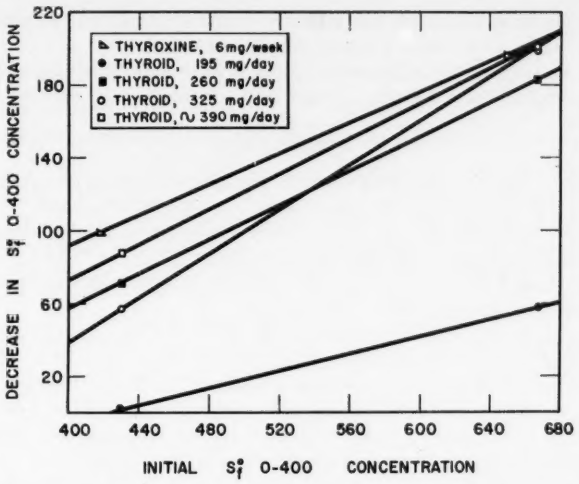

FIG. 4. Effect of *l*-thyroxine and various dosages of desiccated thyroid on S_f° 0–400 serum low-density lipoprotein concentrations at low and high initial S_f° 0–400 concentrations.

by another method that 6 mg. per week of *l*-thyroxine would be approximately equivalent in effect to 325 mg. per day of desiccated thyroid.

Rebounds in concentration above initial levels occurred after cessation of *l*-thyroxine injections in the S_f° 0–12, S_f° 20–100, and S_f° 100–400 lipoproteins and in serum cholesterol. These rebounds were greatest four weeks after stopping *l*-thyroxine. After eighteen weeks without *l*-thyroxine, all lipoprotein and cholesterol concentrations had returned to approximately the initial pre-therapy values.

After stopping the administration of 325 mg. per day of desiccated thyroid, rebounds occurred in the levels of S_f° 0–12 lipoproteins and cholesterol, but not in the levels of S_f° 12–20, S_f° 20–100, or S_f° 100–400 lipoproteins. The rebounds were greatest three to six weeks after stopping administration of thyroid which was about the same length of time required for the maximal rebound after cessation of *l*-thyroxine therapy.

It is interesting that there was no rebound in S_f° 12–20 concentration after administration of either *l*-thyroxine or desiccated thyroid, in spite of the large decreases in the concentration of S_f° 12–20 observed during the administration of either substance.

Acknowledgments

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OCCURRENCE OF THYROID NODULES IN CHILDREN FOLLOWING THERAPY WITH RADIOIODINE FOR HYPERTHYROIDISM*

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ABSTRACT

Between May 1945 and July 1953, 213 patients with toxic diffuse goiter were treated with I^{131} at the University of California Hospital, San Francisco. Five to ten years following therapy, thyroid nodules appeared in 3 of the 18 patients less than 20 years old. Two of these children with late nodules were among the 5 treated before the age of 10 years. The evidence suggests that these nodules are true neoplasms. One showed invasion through its capsule and was classified as an invasive adenoma or low-grade carcinoma. Among the 195 radioiodine-treated patients who were over 20 years of age, there were 2 in whom thyroid nodules were palpable at four and six months after euthyroidism had been attained. In these 2 patients the nodules were found at twelve and fifteen months, respectively, after the initial treatment with radioiodine. In no adult was a nodule first discovered at a later time.

ALTHOUGH at the present time radioiodine therapy is rarely employed in the treatment of uncomplicated hyperthyroidism in young patients, radioiodine was used during an earlier period at the University of California Hospital for the treatment of several children with Graves' disease. Between May 1945 and July 1953, 213 patients with toxic diffuse goiter were treated with I^{131}. Included were 5 patients less than 10 years of age at the time of therapy. In 2 of them, after eight and ten years respectively, palpable thyroid nodules developed in glands previously free of such nodules. In 1 of 13 patients between 10 and 20 years of age, palpable thyroid nodules developed five and a half years after therapy with radioiodine. The present report deals primarily with the 3 patients, aged 5, 9 and 17 years, in whom thyroid nodules developed five or more years after treatment with I^{131}.

CASE REPORTS

Case 1

A 5½-year-old girl was first observed in June 1947 with a three-month history of excessive food intake, heat intolerance, increased perspiration, and irritability. When

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asleep, her pulse was 124 per minute. The blood pressure was 150/75 mm. Hg. She had a pronounced fine tremor and her skin was warm and moist. The skeletal age was 7 years. The serum protein-bound iodine (PBI) level was 9.5 μg. per 100 ml. and the basal metabolic rate (BMR) was +26 per cent. Treatment with propylthiouracil (50 mg. per day) for two months resulted in pronounced clinical improvement. In August 1947, a dose of 2 millicuries (mc.) of I^{131} was given orally; at this time the thyroid was slightly enlarged but contained no palpable nodules. The patient was considered to be euthyroid within two months after therapy. In November 1949 the serum PBI level was 6.4 μg. per 100 ml. and the five-hour thyroidal I^{131} uptake was 51 per cent. The patient was sleeping well and gaining weight and strength. The thyroid was not palpable and there was no tremor. However, the pulse was 112 per minute and she was still hyperkinetic. The patient was seen at regular intervals thereafter and on clinical examination her thyroid function seemed to remain in the upper range of normal.

In March 1950 the three-hour thyroidal I^{131} uptake was 37 per cent. At that time the serum PBI level was 6.0 μg. per 100 ml., and in June 1951 it was 7.4 μg. per 100 ml.

In October 1956 the thyroid was normal on palpation. In July 1957, ten years after radioiodine therapy, the patient (now 15 years old) returned for re-examination because, during the previous two weeks, her mother had noted a nodule in the right side of the neck. At this time the patient was nervous, hyperkinetic, and had mild heat intolerance and a fine tremor. However, she was gaining weight, her skin was normal, and there were no eye signs. The pulse was 96 per minute and the blood pressure was 135/80. The thyroid was about one and a half times normal size and contained palpable nodules. One, in the right lobe, was estimated to measure 2 cm. in diameter. The five-hour thyroidal I^{131} uptake was 45 per cent and the serum PBI level was 6.8 μg. per 100 ml.

Subtotal thyroidectomy was performed on August 24, 1957. At operation, the left lobe did not contain any palpable nodules but did show evidence of radiation injury, manifested by adherence to the sternothyroid muscle and a firm scarred lobe. The right lobe was much larger than the left and contained 2 nodules. The right lobe, isthmus, and a small portion of the left lobe were removed, leaving the posterolateral capsule.

The material removed consisted of a segment of thyroid tissue measuring 5×2×2 cm.; it contained 2 encapsulated nodules, each measuring approximately 2 cm. in diameter. One was cystic and contained brownish fluid; the other was solid. Each nodule was circumscribed and encapsulated. Both nodules had microfollicular and macrofollicular patterns. In the solid nodule the thyroid follicles lay in an edematous, mucoid stroma and only the larger follicles contained colloid. In the second nodule some portions were trabecular, although the central portion was degenerated, hemorrhagic and cystic. The cells of both nodules were uniform; the nuclei were round or oval and contained moderately abundant, finely dispersed chromatin. Cytoplasm was abundant, finely granular, and eosinophilic. Mitotic figures were not observed in these nodules. Many smaller but similar nodules were present throughout the remaining parenchyma (Fig. 1).

Between nodules were segments of residual thyroid parenchyma displaying lobular and follicular atrophy which was probably secondary to irradiation (Fig. 2). The remaining parenchymal cells had large pleomorphic and vesicular nuclei with abundant eosinophilic cytoplasm. The residual lobules were infiltrated with chronic inflammatory cells and showed considerable perifollicular fibrosis (Fig. 2).

The nodules in this patient were classified as follicular adenomas.

Case 2

A 9-year-old girl, first seen in July 1949, had a six-month history of increased prominence of the eyes and pronounced increase in height. As noted by her mother, the pa-

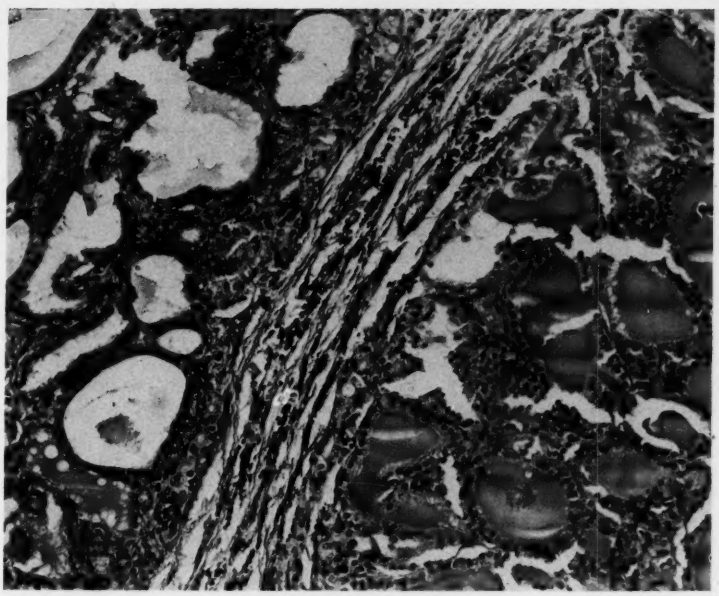

FIG. 1

FIG. 2

FIG. 1. (Case 1). Two adjacent follicular adenomas separated by fibrous capsules. ×100. Hematoxylin and eosin.

FIG. 2. (Case 1). Thyroid parenchyma showing fibrous and lymphocytic infiltration. Few thyroid epithelial cells remain. ×200. Hematoxylin and eosin.

tient was irritable and had increased perspiration, palpitation and weight loss in spite of an excessive appetite. The child was 61 inches tall and weighed 70 pounds. Her skin was warm and moist and she appeared restless. The pulse was 120 per minute and the blood pressure was 130/68. The thyroid gland was approximately one and a half to two times normal size and was smooth and non-nodular. The serum PBI level was 9.6 μg. per 100 ml. and the BMR was +62 per cent.

In August 1949, a dose of 1.2 millicuries of I^{131} was given orally. Within three months the patient had gained 8 pounds and her nervousness, irritability and tremor had disappeared. The pulse rate was now 80 per minute and the skin had become normal. The serum PBI level was 4.5 μg. per 100 ml. Thereafter, rapid growth and moderate elevation of the pulse persisted. In December 1950, the pulse was 100 per minute, serum PBI level 6.7 μg. per 100 ml., BMR +17 per cent, and three-hour thyroidal I^{131} uptake 27 per cent. An additional 0.5 millicurie of I^{131} was then given. The patient was again thought to be euthyroid, although the pulse and growth rate tended to remain elevated.

In July 1953, a thyroid nodule measuring 1.0 cm. in diameter was palpated in the upper pole of the right lobe. Because of recurrence of thyrotoxicosis, the patient was retreated with I^{131} (3.7 millicuries). She then became definitely and permanently euthyroid and the nodule disappeared. In July 1956, other than the fact that the patient was 6 feet tall, no abnormality was observed. The thyroid was barely palpable. She was active physically and scholastically, and was in the top 5 per cent of her senior class in high school.

In August 1957, eight years after the initial therapy with radioiodine, a nodule 2 cm. in diameter was visible and palpable in the right lobe of the thyroid gland. Subtotal thyroidectomy was performed at this time. The left lobe appeared normal except for the effects of irradiation. The right lobe was enlarged, and at operation 2 nodules were seen. The right lobe, isthmus, and a small portion of the left lobe were removed.

The removed tissue proved to contain 3 encapsulated, soft, yellow, opaque nodules. These nodules had similar histologic patterns, which were mainly microfollicular but in part were alveolar and trabecular. The follicles contained colloid, either deeply and uniformly staining, or pale and vacuolated or even mucoid. The cells of these nodules were moderately pleomorphic and had oval or round nuclei containing delicate particles of chromatin. Portions of the nodules showed many mitotic figures. The cytoplasm of the cells was relatively sparse, finely granular and eosinophilic, or distinctly vacuolated (Fig. 3). The nodules were well circumscribed and encapsulated for the most part. However, in one nodule, the thyroid epithelial cells had perforated a thick fibrous capsule and had formed a mushroom-shaped segment which extended to the resected margins of the tissue (Fig. 4). The extruded segment appeared circumscribed and there was no evidence of vascular or lymphatic invasion.

The residual thyroid parenchyma adjacent to the nodules displayed a variable pattern. For the most part, the lobules were atrophic and showed pronounced perilobular and perifollicular fibrosis. The parenchyma was moderately infiltrated with chronic inflammatory cells. The parenchymal cells in the atrophic lobules were pleomorphic and lay in small groups of poorly defined follicles which were usually devoid of colloid. Many cells had irregular, large nuclei and many of them were characterized by abundant eosinophilic cytoplasm (Fig. 5).

Some of the lobules had the pattern of Hashimoto's disease and were infiltrated by lymphocytes and plasma cells. In these lobules the epithelial cells lay in small groups or formed small follicles and had abundant eosinophilic cytoplasm and bizarre pleomorphic nuclei (Fig. 6). Throughout the residual parenchyma were a number of small circumscribed nodules consisting of proliferating epithelial cells which had a microfollicular pattern closely resembling the larger nodules already described.

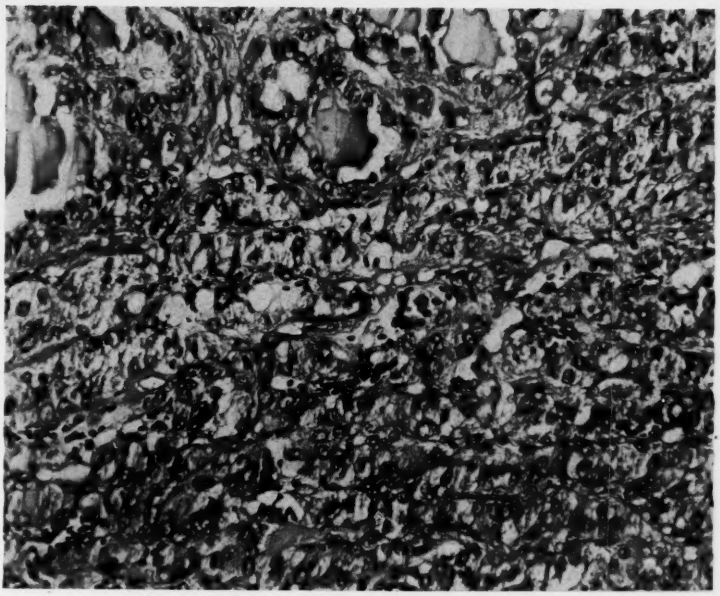

FIG. 3

FIG. 4

FIG. 3. (Case 2). Segment of invasive adenoma. The cells are mildly pleomorphic and have mainly a microfollicular pattern. ×200. Hematoxylin and eosin.

FIG. 4. (Case 2). Margin of thyroid nodule showing invasion of thyroid epithelium through capsule at left. ×40. Hematoxylin and eosin.

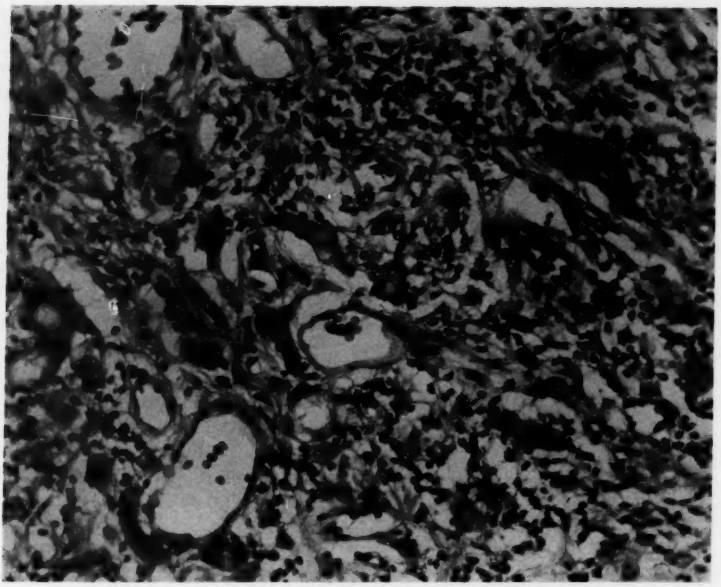

FIG. 5

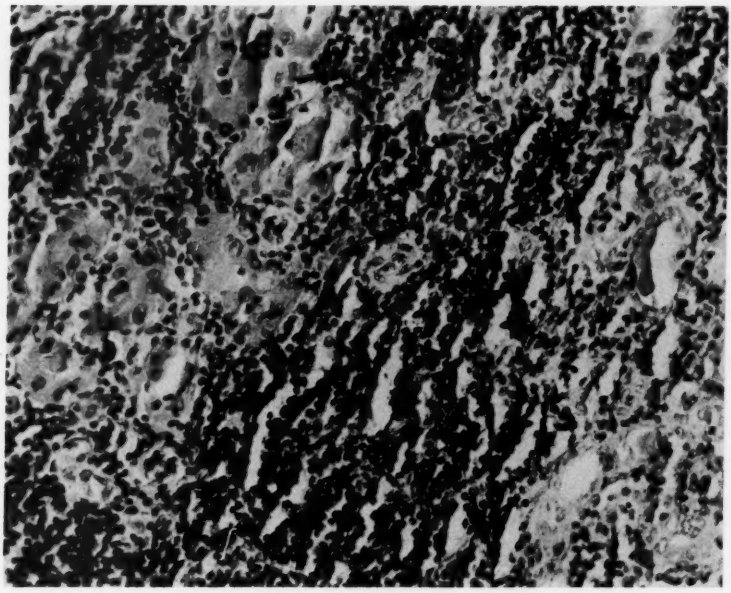

FIG. 6

FIG. 5. (Case 2). Thyroid parenchyma showing follicular atrophy, perilobular fibrosis and lymphocytic infiltration. ×200. Hematoxylin and eosin.

FIG. 6. (Case 2). Thyroid parenchyma showing pattern of Hashimoto's disease. The thyroid epithelial cells have abundant eosinophilic cytoplasm. ×200. Hematoxylin and eosin.

Alpha naphthol esterase (Gomori (1)) and naphthol AS esterase (Pearse (2)) activities were moderate in the nodules, especially in the cells of the microfollicles; less activity was evident in the portions of the nodules showing a trabecular pattern. Greater activity of these esterases was observed in the remaining parenchymal cells of the fibrotic thyroid gland.

Two of the 3 larger thyroid nodules were classified as follicular adenomas. The third, displaying invasion and perforation of the capsule, was classified as an invasive adenoma or a low-grade follicular carcinoma.

Case 3

A 17-year-old girl, first seen in January 1948, had a typical history of thyrotoxicosis of twelve months' duration. The pulse rate was 96 per minute and the blood pressure was 125/82. A fine tremor and warm moist skin were noted. The thyroid was symmetrically enlarged to about twice normal size and no nodules were palpable. The serum PBI level was 8.6 μg. per 100 ml. and the 24-hour thyroidal I^{131} uptake was 72 per cent. A dose of 3.6 millicuries of I^{131} was given orally. Within three months the patient was euthyroid and the thyroid gland was barely palpable.

The patient continued to be well until July 1953, when after an absence of one year, she returned for re-examination because she had recently discerned a lump in her neck. The patient was euthyroid. A nodule 2 cm. in diameter was visible and palpable just to the left of the thyroid isthmus. A subtotal thyroidectomy was performed. The removed tissue consisted of 2 thyroid lobes weighing a total of 6.5 Gm. The right lobe contained a soft, pale, fatty, circumscribed nodule measuring 3.5 cm. in diameter. The left lobe contained a 1-cm. nodule and several small nodules of similar appearance. The remaining parenchyma was firm, pale and pink.

The nodules were circumscribed and separated from the rest of the thyroid gland by thick fibrous capsules. They had follicular and trabecular patterns and mildly pleomorphic cells (Fig. 7). The nuclei varied slightly in size, and the majority contained coarse particulate chromatin. In one nodule the cells were of moderate size and contained little cytoplasm. In the smaller nodule in the left lobe, the cells were larger and had a granular, vesicular cytoplasm. There were few mitotic figures.

The remaining thyroid parenchyma contained numerous small nodules of varying size, which had microfollicular or macrofollicular patterns. The lobules were distorted. The cells of the lobules were pleomorphic and had large hyperchromatic nuclei and abundant eosinophilic cytoplasm. Some portions of the supporting stroma were heavily infiltrated by lymphocytes.

The nodules in this patient were classified as follicular adenomas.

RESULTS AND DISCUSSION

Of the 213 patients treated with I^{131} for toxic diffuse goiter, 170 had had no previous thyroid surgery and 43 had thyrotoxicosis recurrent after subtotal thyroidectomy (Table 1). In only 2 of the 195 patients more than 20 years old were nodules known to have developed one year or more after beginning therapy; in these 2, the nodules were discovered at twelve and at fifteen months after the initial treatment with I^{131}. However, in 2 of 5 patients who were less than 10 years old, and in 1 of 13 patients between 10 and 20 years old, such nodules did develop. All patients were observed closely. Follow-up observations on those unwilling or unable to

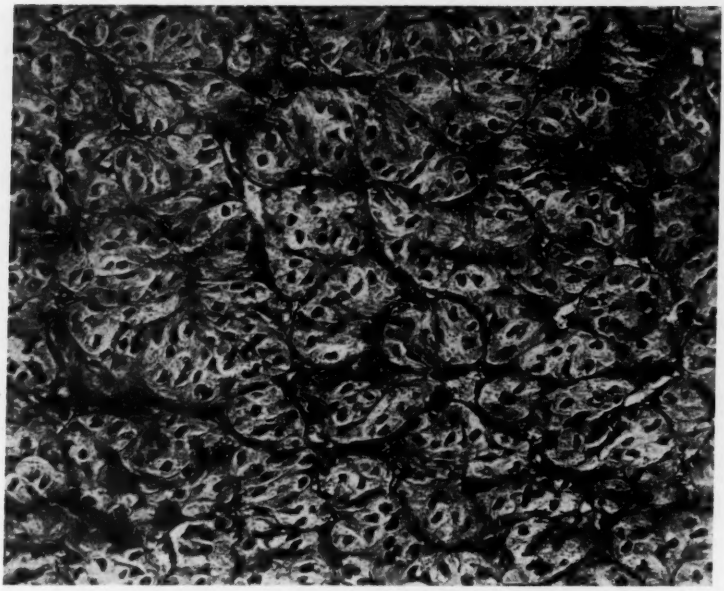

FIG. 7. (Case 3). Trabecular pattern in adenoma. The cells are mildly pleomorphic. ×200. Hematoxylin and eosin.

return for re-examination were obtained from their personal physicians or by direct questionnaire. The thoroughness of the follow-up observations is probably about the same for both the adult and the younger groups. The nodules in the 3 patients less than 20 years of age were readily visible; in fact, 2 of these patients returned for unscheduled visits because of an enlargement of the thyroid gland that was visible to the patient or the family. It is unlikely that similar lesions in the adult were missed.

Although thyroid nodules in the adult may become palpable at the time of completion of therapy with radioiodine, the appearance of nodules after a longer period is unusual. Reports of large groups of patients treated with radioiodine by Chapman and Maloof (3) and by Clark and Rule (4) do not include descriptions of late nodules. In 2 of 275 patients over 20 years of age with toxic diffuse goiter treated by Larsson (5), small nodules developed about two years after beginning therapy. The ages of these 2 patients were not given.

In the current series, 1 of the 2 adults with nodules palpable twelve months or more after beginning therapy with I^{131}, was a 28-year-old woman whose thyroid gland before therapy was estimated to be four to five times normal size. Firm involutionary areas but no definite nodules were pal-

pable. Her thyroid shrank to approximately normal size after therapy. Nodules became palpable six months after she became euthyroid. Histologically, these were interpreted as small macrofollicular adenomas. The second patient was a 63-year-old woman whose thyroid was enlarged to two or three times normal size. A total of 15 millicuries of I^{131}, given in three doses, was required to control increased thyroid function. The gland decreased only from an estimated 45 Gm. to 35 Gm., although most toxic diffuse goiters readily shrink to a normal size or smaller. The nodules in this patient were noted within four months after the last administration of radioiodine. It is possible that these nodules were present before therapy and were disclosed by shrinkage of the tissue induced by the radiation.

Nodules in the patients aged 5, 9 and 17 years did not become palpable until five and a half to ten years after initiation of radioiodine therapy. Since in all 3 patients there was a period during which the gland was of normal size and without nodules, it might be concluded that the nodules were of neoplastic origin. Further, it would appear that, after therapy with radioiodine, the thyroid glands of children are more likely to form such nodules than are those of adults. There is evidence in the literature, from studies both in human subjects and animals, that the thyroids of youths and of adults may respond differently to certain stimuli. The thyroid glands of infants and children (6–9) may be the site of development of carcinoma after irradiation of the head, neck or thymus gland, whereas there is no convincing evidence that the adult human thyroid gland (at least as found in toxic diffuse goiter) responds to irradiation in a similar manner (9–11).

TABLE 1. LATE APPEARANCE OF THYROID NODULES IN 213 PATIENTS TREATED WITH I^{131} FOR TOXIC DIFFUSE GOITER

Age (yrs.)	No. of patients with diffuse goiter		No. of patients with post-surgical recurrence	
	total	with nodule after 1 year	total	with nodule after 1 year
<10	5	1 at 10 yrs. 1 at 8 yrs.	0	0
10–20	11	1 at 5½ yrs.	2	0
>20	154	1 at 1 yr. 1 at 1¾ yrs.	41	0
Total	*170*	*5*	*43*	*0*

Doniach (12) has suggested that the apparent difference in sensitivity of the thyroid gland of infants and adults, as indicated by carcinogenesis, results from the fact that the thyroid gland of the infant will subsequently undergo considerable mitotic activity coincident with an increase in the number of thyroid cells. The adult thyroid gland, however, normally undergoes little cellular replacement. In support, Doniach has shown that the benign and malignant neoplasms that develop in the irradiated thyroid of the adult rat, occur in increased numbers when cell division is additionally stimulated by administration of goitrogens. Lindsay, Potter and Chaikoff (13) have shown that the highest incidence of neoplasms induced in the thyroid of the rat injected with I^{131} occurs with a relatively small dose (25 microcuries). In their study, the number of induced neoplasms decreased as the dose of radioiodine was increased and as the destructive effects of the irradiation became more pronounced. These authors concluded that the benign and malignant neoplasms that appeared in the thyroid gland of the rat irradiated with radioiodine originate as foci of nodular hyperplasia and regeneration. They also concluded that the carcinoma induced in the irradiated rat might develop in a previously benign adenoma. In children who have received irradiation therapy as compared with those who have not, Simpson, Hempelmann and Fuller (14) found a greater incidence of thyroid adenoma as well as carcinoma. In the presently reported Case 2, the simultaneous appearance of benign nodules and of one nodule that probably was of low-grade malignancy may be an example of a neoplastic nodule progressing from the benign to the malignant state.

Whatever the explanation, the present study establishes that nodular hyperplasia of the thyroid may occur if functional thyroid tissue is left in the young patients treated with radioiodine for toxic diffuse goiter. In fact, invasive changes suggestive of early malignancy resulted in one instance. These findings, although based on a small series, seem to provide an adequate rationale for not using radioiodine in the treatment of patients who are less than 20 years of age. If, in this age group, special circumstances make therapy with radioiodine the treatment of preference regardless of age, it is perhaps safer to give doses of radioiodine sufficiently large to produce hypothyroidism and thus reduce the possibility of leaving thyroid tissue capable of undergoing subsequent diffuse or nodular hyperplasia. The patient would then be given thyroid as replacement therapy.

Acknowledgment

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EXOPHTHALMIC GOITER IN CHILDREN*

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ABSTRACT

A total of 253 children less than 15 years of age who had exophthalmic goiter have been seen at the Mayo Clinic during the 48-year period, 1908 through 1955. There were 217 girls and 36 boys, a ratio of about 6:1. Observations in this series indicate that the disease in children does not appear to differ significantly from that in adults, although there is less tendency for children to have severe exophthalmos. Exophthalmos that occurs during childhood may persist for years after thyrotoxicosis has been relieved. Diabetes mellitus is observed more frequently among children with exophthalmic goiter than would be expected as a chance occurrence. Exophthalmic goiter during childhood, if adequately treated, does not alter adolescent development. Subtotal thyroidectomy is a satisfactory and safe method of treating exophthalmic goiter in children. There were only 4 hospital deaths among 196 children treated by this means. No surgical death has occurred since 1933 among children treated by this method. Recurrence of hyperthyroidism has not been eliminated by more extensive resection of the thyroid gland. Myxedema, although not considered a serious complication, occurs in a high percentage of patients treated by the extensive subtotal resection of the thyroid gland that is now carried out at this clinic. Hypoparathyroidism, keloid formation in the operative scar, and injury to the recurrent laryngeal nerves rarely occurred in the patients of this series. It is our opinion that a final evaluation of the best method of treating exophthalmic goiter in children must await an extensive experience with the other methods of treatment.

EXOPHTHALMIC goiter in children has been the subject of a number of publications from the Mayo Clinic (1–3). Several investigators have presented excellent reviews of the clinical and laboratory manifestations of this disease in children (4–8). During recent years, considerable controversy has arisen as to whether children with exophthalmic goiter are best treated with antithyroid drugs (9–11) or by surgical subtotal thyroidectomy (12). The present study was undertaken with the hope that a review of our experience over a 48-year period would add to our

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understanding of the natural course of the disease, and more specifically to appraise subtotal thyroidectomy as a method of treating exophthalmic goiter in childhood.

MATERIAL AND METHOD

During the period 1908 through 1955 at the Mayo Clinic, 253 children, all less than 15 years of age, had thyroid disease diagnosed as exophthalmic goiter. The distribution of these patients according to year of diagnosis and sex is presented in Figure 1. The disease has been seen with approximately the same frequency throughout the years except during the decade of 1920 through 1929 when the number of both adults and children increased.

The criteria used in establishing the diagnosis of exophthalmic goiter have been discussed in a previous publication (3) and need not be reviewed in detail. In the years before the advent of more refined technics for the study of thyroid function the diagnosis was established on the basis of the clinical history, physical examination, and course of the disease. Since the advent of radioactive iodine and methods for measuring the protein-bound iodine of the blood, the diagnosis has been confirmed by one or the other of these methods in most instances. The basal metabolic rate has been of help in confirming the diagnosis and in following the course of the disease in most of the patients.

In each case included in this study the diagnosis was confirmed by more than one observer. The records of the patients were reviewed and an effort was made to follow each patient by questionnaire. In most instances information was obtained from the patient by letter but in some instances it was obtained from members of the family or from physicians who attended the patient during more recent years. Some of the patients continued to return to the clinic for care. Data on the duration of the follow-up are

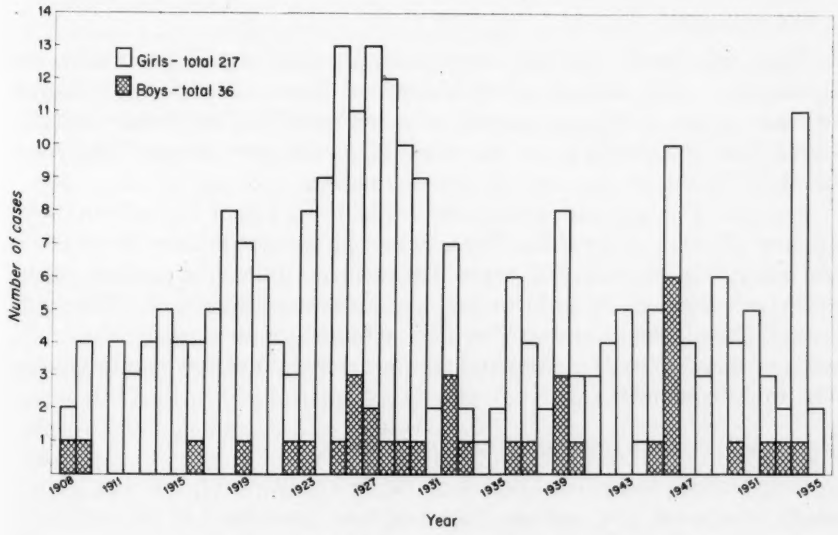

FIG. 1. Exophthalmic goiter in children. Distribution by year of diagnosis and by sex.

TABLE 1. EXOPHTHALMIC GOITER IN CHILDREN. FOLLOW-UP PERIODS AND STATUS OF PATIENTS IN 253 CASES

Years of follow-up	Patients		
	Living	Dead	Total
Less than 1	12	12	24
1– 4	29	7	36
5– 9	36	4	40
10–14	40	3	43
15–19	27	2	29
20–24	12	1	13
25–29	27	1	28
30–34	19	0	19
35–39	11	1	12
40–44	7	0	7
45–49	2	0	2
Total	222	31	253

given in Table 1. It should be noted that 60.5 per cent of the group were followed for ten or more years since the diagnosis of exophthalmic goiter was established. Patients known to have died from any cause are represented in the table.

OBSERVATIONS

Effect on growth

When adequately treated, exophthalmic goiter of childhood does not significantly alter adolescent development. Thus 147 children followed beyond the age of 19 years presented no evidence that the disease had deterred their development in any way. Many are now married and have families. They have pursued various occupations.

Records of height and weight are available on 138 of the patients who are now 20 years old or older. The average age of these patients is 34 years, the oldest being a man of 57 years. The women attained an average height of 65 inches (range, 60 to 71 inches) and an average weight of 135 pounds (range, 92 to 205 pounds). The men attained an average height of 71 inches (range, 65 to 76 inches) and an average weight of 165 pounds (range, 132 to 230 pounds).

Residual symptoms from previous thyrotoxicosis

In appraising the general health of patients of this study we were particularly interested in symptoms that might be considered as residual from the previous thyrotoxicosis; therefore patients were asked to note specifically prominence of the eyes, nervousness, tremor, excessive sweating or

tachycardia. Of the 147 patients who are now adults, 22 complained of residual prominence of the eyes. In most instances they considered their eyes to be only slightly more prominent than in normal persons, but in 2 the proptosis was severe enough to be disturbing. No patient of this group has manifest malignant exophthalmos of the degree not infrequently encountered among adults. We do not consider changes in the degree of proptosis to be directly related to changes in the degree of hyperthyroidism. Several patients have proptosis more than twenty-five years after having been relieved of thyrotoxicosis.

Thirty-four of the 147 patients who are now adults reported some degree of nervousness, tremor, tachycardia, excessive sweating or palpitation on occasion. These symptoms are difficult to evaluate but cannot be considered as serious, since all except 3 of these patients considered themselves to be in good health.

Associated diseases

It is to be expected that various diseases would occur among patients of this study. This has been the case, but except for diabetes mellitus there does not appear to be more than chance association of these diseases with exophthalmic goiter. Among the diseases noted, in 1 instance each, are psoriasis, tuberculosis, narcolepsy, epilepsy, multiple sclerosis rheumatic heart disease and nephrolithiasis.

Diabetes mellitus is known to have developed in 3 boys and 6 girls. In 4 instances it preceded thyrotoxicosis, and in 5 it appeared within six months to ten years after thyrotoxicosis had been diagnosed. Two patients died in a state of diabetic keto-acidosis. A girl, 12 years of age, was seen at the clinic in 1925 because of exophthalmic goiter. After subtotal thyroidectomy she was euthyroid for fifteen years before Addison's disease developed and she died during a crisis.

Deaths

Thirty-one of the 253 patients are known to have died (Table 2). Ten died in the hospital following operation for relief of thyrotoxicosis—8 at the Mayo Clinic and 2 elsewhere. The 8 hospital deaths at the clinic, all of which occurred before 1934, will be discussed in more detail later. Two patients died of exophthalmic goiter in the hospital—in one case complicated by pneumonia and in the other by heart failure. An additional patient died six months after refusing surgical treatment and returning to her home. We have already noted that 2 patients died of diabetic keto-acidosis and 1 of addisonian crisis. In the remaining 15 patients the cause of death, when we were able to ascertain it, did not appear to be directly related to thyrotoxicosis.

Treatment

The patients of this series may be divided into 3 groups according to the type of treatment rendered at the Mayo Clinic.

Group 1 (not treated surgically at the clinic). This group consisted of 38 patients (33 girls and 5 boys). Eleven of the 38, however, were treated surgically elsewhere; 2 of these are known to have died, one in 1918 and the other in 1929. The remaining 27 patients were not treated surgically for a variety of reasons (Table 3). Two of the 27 died of exophthalmic goiter, as indicated earlier. Two had moderately severe exophthalmos but little evidence of thyrotoxicosis, and it seemed unlikely that thyroidectomy would be of benefit. Propylthiouracil was apparently effective in controlling the disease in 3 patients; therefore surgical treatment was not recommended. Four patients were not treated surgically because of complicating diseases. Two patients were given Lugol's solution initially in preparation for surgical treatment and allowed to go home; they did not return as

TABLE 2. EXOPHTHALMIC GOITER IN CHILDREN. DEATHS FROM ALL CAUSES (31 DEATHS IN 253 CASES)

Year diagnosis made	Age (yrs.) & Sex	Treatment		Comment
		Type	Year	
Deaths in hospital after surgical treatment (8 at Mayo Clinic and 2 elsewhere)				
1915	14 F	Vessels ligated	1915	Very toxic. Died 4th p.o. day
1915	12 M	Vessels ligated. Subtotal thyroidectomy	1915 1916	Very toxic. Died 4th p.o. day
1915	10 F	Vessels ligated. Vessels ligated	1915 1916	Died in hospital (elsewhere) after another operation; word of death, 1926
1919	11 M	Quinine injected into thyroid; vessels ligated	1919	Died 1st p.o. day in crisis
1920	13 F	Boiling water injected into thyroid	1920	Died 23rd p.o. day of hyperthyroidism and heart failure
1921	13 F	Vessels ligated	1921	Died of hyperthyroidism complicated by subac. bact. endocarditis
1925	5 F	Subtotal thyroidectomy	1925	Died after operation; had hyperthyroidism and pneumonia
1929	6 F	Subtotal thyroidectomy	1929	Died 3rd p.o. day (elsewhere)
1931	11 F	Subtotal thyroidectomy	1931	Died 1st p.o. day in crisis
1933	4 F	Subtotal thyroidectomy	1933	Died 1st p.o. day, very toxic; convulsive disorder, brain cyst, osteoporosis

(Continued on next page)

TABLE 2. (*continued*)

Year diagnosis made	Age (yrs.) & Sex	Treatment		Comment
		Type	Year	
Deaths not related to surgical treatment (2 at Mayo Clinic—others elsewhere)				
1908	12 F	Vessels ligated on rt.	1908	Recurrence. Died 1½ yr. after operation; cause?
1909	13 F	Vessels ligated	1909	Good health 13 yrs. after operation; word of death, 1932; cause?
1909	13 F	Vessels ligated	1909	Died 1919 of postpartum hemorrhage
1911	14 F	Vessels ligated	1911	Hyperthyroidism recurred. Died 1912; cause?
1911	14 F	Vessels ligated. Subtotal thyroidectomy	1911 1921	Died 1929, rheum. heart dis., mitral insuff.
1912	14 F	Vessels ligated. Subtotal thyroidectomy	1912 1913	Drowned 1950
1913	13 F	Vessels ligated. Subtotal thyroidectomy	1913 1914–1915	Fair control of hyperthyroidism. Nephritis 1915. Died 1917; cause?
1913	13 F	Vessels ligated. Subtotal thyroidectomy at clinic, also elsewhere	1913 1914	Well for 29 yrs. after second subtot. thyroidectomy Cause of death?
1915	14 F	Vessels ligated. Subtotal thyroidectomy	1915 1915	Seen at clinic in 1923 and was euthyroid. Died 1923; cause?
1916	14 F	Hot water injected. Subtotal thyroidectomy	1916 1917	Died 1921 of cellulitis and pneumonia
1917	11 F	Vessels ligated	1917	Had dementia praecox and encephalitis. Died 1932
1918	14 F			Treatment refused. Died 6 mos. after clinic visit; cause?
1920*	14 F			Died 8 days after admission to hospital—heart failure
1923	14 F	Vessels ligated. Subtotal thyroidectomy	1923 1928	Learned of death in 1955; cause?
1924*	14 F			Died 3 days after hospitalization, of pneumonia and hyperthyroidism
1925	3 M	Subtotal thyroidectomy	1925	Died 10 mos. after operation; diabetic coma
1925	12 F	Subtotal thyroidectomy	1925	Died 1939 of Addison's disease
1926	13 F	Subtotal thyroidectomy	1926	Died 1929 of recurr. exoph. goiter and diabetes m.
1937	11 F	Subtotal thyroidectomy	1937	Postop. myxedema. Died suddenly in 1938 after being frightened
1942	14 F	Subtotal thyroidectomy	1942	Suicide 1953
1946	14 M			Died 1948 of hepatic cirrhosis complicated by diabetes m. and hyperthyroidism

* Died at Mayo Clinic.

TABLE 3. EXOPHTHALMIC GOITER IN CHILDREN: PATIENTS NOT TREATED SURGICALLY AT MAYO CLINIC

Year diagnosis made	Age (yrs.) & Sex	Follow-up (yrs.)	Comment
		Surgical treatment elsewhere	
1919	11 F	36	Operation prior to clinic visit. Good health
1918	14 F	Died	Returned home for operation. Died 6 mo. later; cause?
1924	14 F	32	Operation 1924, 1925. Good health after second operation
1927	13 M	½	Not traced since 1927. Good health 3 mo. postop.
1929	6 F	Died	Died 3 days postop.
1934	13 F	5	Good health 1939. Not traced since 1939
1936	7 F	3	Good health 1939. Not traced since 1939
1936	5 F	19	Good health
1945	12 F	10	Good health. Myxedema treated
1948	14 F	7	Good health
1954	12 F	0	Returned home for operation. Not traced
		Surgical treatment not recommended, or refused by patient	
1929	10 F	10	Exoph. goiter without hyperthyroidism. No progression during 10 yrs.
1949	12 F	7	Exoph. goiter without hyperthyroidism. No progression during 7 yrs.
1949	11 F	8	Successfully treated with propylthiouracil
1954	12 F	0	Dis. controlled with propylthiouracil at time of visit
1954	10 F	1	Dis. controlled with propylthiouracil
1921	12 F	0	Dis. complicated by mental deficiency and psychosis
1924	11 F	15	Mild hyperthyroidism complicated by psychoneurosis 1924. Operation elsewhere 1929. Well 1939
1946	14 M	Died	Dis. complicated by diabetes m. and hepatic cirrhosis. Died 1948
1951	11 F	4	Dis. complicated by chr. ulc. colitis. Controlled with Lugol's sol.
1920	14 F	Died	Died 8 days after admission to hospital—heart failure
1924	14 F	Died	Died 3 days after admission to hospital—pneumonia
1923	13 F	16	Mild hyperthyroidism. Apparently controlled by Lugol's sol.
1924	8 F	31	Mild hyperthyroidism. Apparently controlled by Lugol's sol.
1925	11 F	4	Mild hyperthyroidism
1925	5 F	15	Mild hyperthyroidism
1925	8 F	31	Mild hyperthyroidism
1926	12 F	30	Lugol's sol. for 2 yrs. Nervousness 1956
1927	9 M	12	Mild hyperthyroidism. Apparently controlled by Lugol's sol.
1927	11 F	11	Mild hyperthyroidism. Adrenal disease suspected 1938. Not traced since 1938
1929	11 F	2	Mild hyperthyroidism. Apparently controlled by Lugol's sol.
1930	14 F	25	Mild hyperthyroidism. Apparently controlled by Lugol's sol.
1930	14 M	25	Mild hyperthyroidism. Apparently controlled by Lugol's sol.
1932	10 F	7	Evidence of hyperthyroidism when last seen 1939. Operation refused
1932	10 M	7	Operation suggested. Apparently controlled by Lugol's sol.
1936	11 F	½	Mild hyperthyroidism. Apparently controlled by Lugol's sol.
1946	3 F	0	Operation recommended. Lugol's sol. given. Not traced
1954	9 F	½	Improved while taking Lugol's sol. in preparation for operation. Operation refused

planned, and have not been traced. The remaining 14 patients, all seen before the advent of newer technics for measuring thyroid function, were considered to have mild hyperthyroidism and were treated with iodides and reduction of activities.

Group 2 (treated by miscellaneous surgical procedures). This group consisted of 19 patients (18 girls and 1 boy) treated by such surgical procedures as ligation of the thyroid vessels or injection of sclerosing substances into the

thyroid gland (Table 4). The procedures carried out on these patients were abandoned years ago because they were considered inadequate for the control of the disease; the patients are included in this study only for the sake of completeness. Four patients of this group died after surgical treatment; of these, 2 died following ligation of the thyroid vessels, 1 following ligation and injection of quinine, and 1 following an injection of boiling water into the thyroid gland. Four patients of the group are known to have had additional operative procedures carried out elsewhere; 1 of these died in the hospital in the postoperative period. All patients of this group were seen before the advent of Lugol's solution. They are not considered to

TABLE 4. EXOPHTHALMIC GOITER IN CHILDREN: 19 PATIENTS SEEN BEFORE 1922 AND INADEQUATELY TREATED AT MAYO CLINIC BY SURGICAL MEANS

Year seen	Age (yrs.) & Sex	Operation		Follow-up (yrs.)	Comment
		Year	Type		
1908	14 F	1908	Ligation, rt. sup. thyroid art.	3	Recurred 1911 (treated elsewhere); died; cause?
1909	13 F	1909	Ligation	13	Died 1932; cause?
1909	14 F	1919	Ligation	30	Good health 1939. Not traced 1956
1909	14 F	1909	Ligation	10	Died 1919, complications of pregnancy (postpartum hemorrhage)
1911	14 F	1911	Ligation	1	Died 1912. Poor health after operation here. Cause of death?
1911	8 F	1911	Ligation (2×)	45	Fair health 1956. Housewife
1912	7 F	1912	Double ligation	44	Good health 1956. Housewife
1913	13 F	1913	Ligation	3	Good health 1916. Not traced since 1916
1914	13 F	1914	Ligation	25	Recurred 1932; resection 1932 (elsewhere). Well in 1939, not traced 1956
1914	8 F	1914	Ligation	—	Not traced
1915	14 F	1915	Ligation	Died	Hospital death
1916	10 F	1916	Ligation	—	Died 1916 after operation elsewhere
1916	12 F	1916	Ligation (2×)	—	Not traced
1917	11 F	1917	Ligation	15	Died 1932 of encephalopathy
1918	12 F	1918	Ligation (2×)	38	Recurred 1930; subtotal resection elsewhere. Good health 1956
1919	14 F	1919	Ligation (2×)	—	Not traced
1919	11 M	1919	Quinine injected; ligation	Died	Hospital death 1 day postop.
1920	13 F	1920	Boiling water injected	Died	Died 23rd postop. day in hospital
1921	13 F	1921	Ligation	Died	Hospital death

have had adequate surgical treatment and it is this group of patients that resulted in the high surgical mortality rate that was reported before the year 1922.

Group 3 (treated by surgical subtotal thyroidectomy). This group consisted of 196 patients, all of whom were treated by surgical subtotal thyroidectomy. Here it should be said that it is most difficult to appraise any type of treatment of exophthalmic goiter in children, since the disease is of

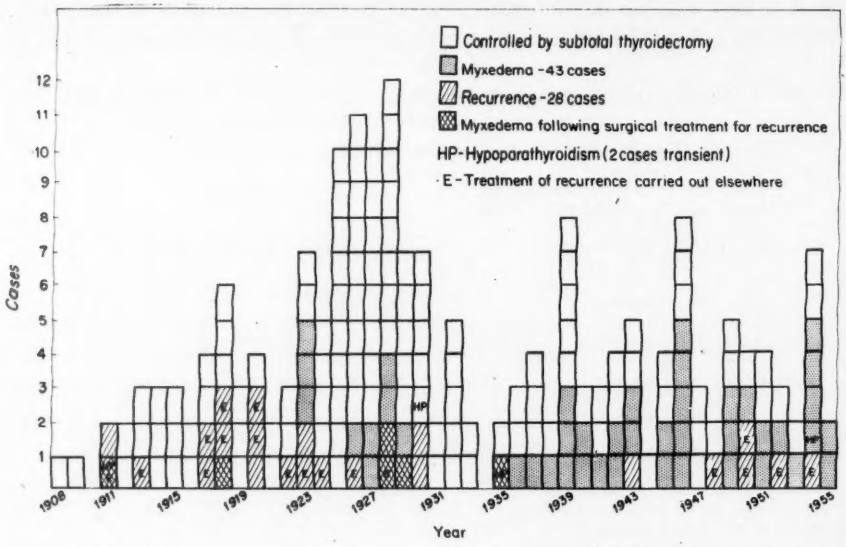

FIG. 2. Exophthalmic goiter in children. One hundred and ninety-six patients treated by subtotal thyroidectomy. Distribution by year of surgical treatment and by complications.

unknown cause, infrequent occurrence and variable severity. No effort is made to assess the results of medical versus surgical treatment. A study of 196 children treated by subtotal thyroidectomy should, however, afford a base line for future comparison with other methods of management. The complications of subtotal thyroidectomy in the treatment of exophthalmic goiter in children include surgical mortality, recurrence of thyrotoxicosis, myxedema, hypoparathyroidism, keloid formation in the operative scar, and injury to the recurrent laryngeal nerves. Figure 2 shows the distribution of the patients according to year of surgical treatment and information relative to the more important complications. Not indicated in the figure is the fact that 25 patients of the group were treated by preliminary ligation of the thyroid vessels followed by subtotal thyroidectomy. In 5

cases sclerosing substances were injected into the thyroid gland at the time of ligation of the vessels.

Two patients had previously been subjected to surgical treatment elsewhere, six and eight years respectively before their visits to the clinic. Twelve additional patients had been treated unsuccessfully with antithyroid drugs. In most instances the antithyroid drugs had been administered in inadequate amounts or for too short a period to expect control of the disease. In a few instances the administration of antithyroid drugs was discontinued because of increase in size of the goiter. Six patients had previously been treated unsuccessfully by irradiation of the thyroid gland—3 with roentgen rays, 2 with radium, and 1 with radioiodine.

All the patients in this series seen since 1922 were treated with iodine in preparation for surgical treatment. Several patients were treated surgically after having been treated unsuccessfully with Lugol's solution for periods of six or more months.

Four hospital deaths occurred among the 196 patients treated by subtotal thyroidectomy. A 12-year-old boy seen at the clinic in 1916 because of exophthalmic goiter complicated by asthma was treated by ligation of the thyroid vessels followed by subtotal thyroidectomy. In the postoperative period he had respiratory stridor which was not relieved by tracheotomy and he died. Death was attributed to edema of the glottis and to thyrotoxicosis. Two girls, seen in 1925 and 1931 at the ages of 5 and 11 years respectively, died of respiratory failure on the first postoperative day. In each instance pulmonary edema was considered the cause of death. The fourth death was that of a $4\frac{1}{2}$-year-old girl seen at the clinic in 1933 because of exophthalmic goiter complicated by osteoporosis and multiple fractures. Soon after operation she had convulsions and died. Postmortem examination disclosed a large arachnoid cyst of the brain. No deaths followed surgical treatment of exophthalmic goiter among the 95 children treated by this method at the clinic since the year 1933.

Twenty-eight of the 196 patients treated by subtotal thyroidectomy are known to have had recurrence of hyperthyroidism of such severity as to require further definitive treatment. Seven additional patients who manifested symptoms of mild hyperthyroidism were returned to good health by the administration of Lugol's solution. The 2 patients previously mentioned who had been treated surgically before admission to the clinic are not included among the 28 patients with recurrence, since definite information as to their earlier treatment is not available. In Table 5 information is listed on the interval between the original surgical treatment and the treatment for recurrence in the 27 patients for whom the date of the second period of treatment was definitely known. It is seen that 5 patients required further treatment for hyperthyroidism within one year after sub-

total thyroidectomy. This raised the question of the effectiveness of the operative procedure in control of the disease of these patients. We consider them to have had recurrences rather than uncontrolled hyperthyroidism, since each patient was considered euthyroid at some time after the original operation. Perhaps of greater interest is the fact that 3 patients had recurrence more than twenty-five years after their initial treatment. The average interval between initial treatment and treatment of the recurrence of hyperthyroidism was slightly more than eight years. This fact is most disturbing and prompts one to ask if the second episode does not indeed rep-

TABLE 5. RECURRENCE AFTER SUBTOTAL THYROIDECTOMY FOR EXOPHTHALMIC GOITER IN CHILDREN. CASES ACCORDING TO INTERVAL BETWEEN ORIGINAL SURGICAL TREATMENT AND TREATMENT FOR RECURRENCE

Years after original treatment	No. of cases
Less than 1	5
1– 4	6
5– 9	6
10–14	5
15–19	2
20–24	0
25–29	3
Total	*27**

* In 1 additional case, date of treatment for recurrence is not known.

resent a new attack of the disease rather than a recurrence of the old one. Certainly it is evident that in some patients the potential for hyperthyroidism persists for a long time, perhaps for life. Even more disturbing is the fact that 2 of the 28 patients with recurrence each had 3 subtotal thyroidectomies for hyperthyroidism over periods of seven and twenty-four years respectively, and a third patient had 4 subtotal thyroidectomies over a period of eleven years. Ten of the patients with recurrence had been treated by ligation of the thyroid vessels preliminary to their first subtotal thyroidectomy. Eight of these ligations were carried out at the Mayo Clinic and 2 elsewhere.

The treatment used in the control of recurrent hyperthyroidism consisted of a second subtotal thyroidectomy in 22 of the patients—10 at the Mayo Clinic and 12 elsewhere; the remaining 6 patients were all treated elsewhere, 3 with radioactive iodine and 3 with antithyroid drugs.

Myxedema followed subtotal thyroidectomy in 49 patients. In 6 the myxedema was not present until after a second or third subtotal thyroidec-

tomy. Two of these patients had surgical treatment for recurrence elsewhere, and the remaining 4 were treated at the clinic. Thus in 47 patients, myxedema developed as a result of surgical treatment given exclusively at the clinic. In each instance the myxedema was successfully treated by the administration of desiccated thyroid. It is interesting that in some cases myxedema did not become manifest for many years after surgical treatment, but in most instances it was apparent within three to six months after operation.

Transient hypothyroidism of varying degrees of severity was observed among many of the patients of this study but did not require treatment. As more extensive surgical resections of the thyroid have been carried out, the incidence of myxedema has increased. It is disturbing to note that the incidence of recurrence of hyperthyroidism has not decreased in spite of the more extensive surgical procedures.

In our opinion there is no foundation for the often expressed concern about the varying needs of thyroid hormone during puberty. Growth and development proceed normally in youngsters with myxedema if they are provided with an adequate amount of thyroid substance. We do not consider myxedema a serious complication of subtotal thyroidectomy but we are mindful of the fact it requires consideration in evaluating this form of treatment for exophthalmic goiter. A child with myxedema requires medical supervision and daily medication.

Transient hypocalcemia manifested by increased neuromuscular irritability was observed occasionally during the first days after subtotal thyroidectomy but rarely was severe enough to require treatment. True hypoparathyroidism was observed twice among the children of this group subjected to subtotal thyroidectomy. In one case hypoparathyroidism followed subtotal thyroidectomy after previous treatment by ligation of the thyroid arteries. In the second case the condition followed a second subtotal thyroidectomy which was carried out elsewhere because of recurrent hyperthyroidism. Two additional patients, seen in 1935 and 1954, have described symptoms suggesting mild hypoparathyroidism but in each case the relief afforded by small amounts of medication makes the diagnosis of hypoparathyroidism most unlikely.

Keloid formation was occasionally seen in the operative scar. It was serious enough in 7 patients to justify roentgen or radium irradiation. Although difficult to appraise, it should be realized that the presence of an operative scar of the neck is frequently a matter of concern to adolescent girls and to their parents.

Injury to the recurrent laryngeal nerves has not been a problem among the children treated surgically for exophthalmic goiter at the Mayo Clinic. It has long been our practice to examine the vocal cords before and

after thyroidectomy. In 1 patient, seen in 1924, paralysis of a vocal cord was noted in the postoperative period; this patient has had no symptoms referable to this paralysis during the intervening years. Another patient, seen in 1954, had transient paralysis of one vocal cord, which subsided spontaneously after three or four months.

Pathologic features

The pathologic changes in the thyroid gland of patients seen during the 25-year period, 1931 to 1955 inclusive, were reviewed; the majority of surgical specimens removed prior to 1931 were no longer available for study. Two to 4 blocks were cut from each gland, thus ensuring adequate histologic sampling. The characteristic histologic changes of Graves' disease, including increase in cell height, epithelial hyperplasia, and thinning of colloid with scalloping of margins, were seen in all cases, but these changes varied greatly in degree. In general, severely toxic hyperthyroidism was associated with marked hyperplasia, and clinically mild hyperthyroidism with slight or focal areas of hyperplasia. However, no constant correlation could be made between severity of symptoms and histologic findings, since prior treatment with Lugol's solution or antithyroid drugs, or both, frequently produced alteration in the typical histologic structure.

Some degree of lymphocytic infiltration was seen in approximately 60 per cent of cases. This change was usually focal in character and minimal in amount. In only 5 per cent of cases was moderate or marked, fairly diffuse lymphocytic thyroiditis noted throughout the gland. No oxyphilic epithelium or evidence of "exhaustion atrophy" was observed.

The development of postoperative myxedema could not be correlated with the histologic findings in the gland with particular reference to "thyroiditis." In many instances postoperative myxedema occurred in patients whose glands showed marked hyperplasia and little or no lymphocytic infiltration.

One patient was found to have a small papillary carcinoma of the thyroid. The tumor was of low-grade malignancy and measured 8 mm. in diameter.

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MEDULLARY (SOLID) CARCINOMA OF THE THYROID—A CLINICOPATHOLOGIC ENTITY*

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ABSTRACT

Medullary carcinoma is characterized by a solid nonfollicular histologic pattern, the presence of amyloid in the stroma, and a high incidence of lymph node metastases. The tumor is clinically of an intermediate rather than a high grade of malignancy, despite its undifferentiated character. It is important to distinguish it from the highly malignant solid form of anaplastic carcinoma, and from papillary carcinoma, which is of a lower grade of malignancy.

Twenty-one cases are reported. Fourteen patients were women and 7 were men; the ages ranged from 33 to 66 years with a median of 50 years. The regional cervical lymph nodes contained metastases at the time of initial surgery in 10 of 21 cases (48 per cent). Distant metastases occurred in 8 patients and were principally evident in the lungs, liver and bone. Amyloid, though of variable amount, was present in the stroma of all tumors of this series. The course of the disease was usually chronic; 12 patients survived six to twenty-seven years after the initial thyroid operation. Eleven patients died (6 because of neoplasm) four months to twenty-seven years after definitive surgical treatment.

THE multitude of histologic variants of carcinoma of the thyroid has resulted in the development of a highly complicated terminology; therefore, the addition of a new diagnostic term should be recommended only upon established necessity. In an appraisal of thyroid carcinoma several years ago, it became evident that a type of tumor existed which apparently had not received previous special recognition, although it had specific clinical and pathologic features. This tumor was basically of solid histologic structure and of nonpapillary configuration, yet had a high potentiality for metastasizing to lymph nodes. At first, because of a suggestive papillary arrangement of cells associated with artifacts in the old formalin-fixed material, it was thought that certain of these neoplasms might be a form of papillary carcinoma; but recent and freshly fixed tissue has yielded

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evidence that the structure is not basically papillary. Formerly, the undifferentiated character of the tumor led to its being included with the group of highly malignant anaplastic carcinomas, yet the clinical behavior indicates that it is of a significantly lower grade of malignancy and of chronic course.

It is the purpose of this report to present the pertinent findings in 21 cases of this medullary or solid form of thyroid carcinoma and to recommend that it receive recognition as a clinicopathologic entity.

MATERIAL AND METHODS

The pathologic material from 600 carcinomas of the thyroid removed at the Cleveland Clinic Hospital, between the years 1926 and 1957, was reviewed. Twenty-one carcinomas proved to be of the particular character considered in this report. The specimens in the early years were formalin-fixed; more recently, tissue was fixed both in formalin and in Zenker's fluid. Sections were stained by the standard hematoxylin-eosin and the hematoxylin-eosin and methylene blue stains, and also by the crystal violet technic for amyloid. Silver stains for demonstration of argentaffin granules were employed in sections of several tumors.

PATHOLOGIC FEATURES

Grossly the neoplasms were of hard consistence, usually white or gray-white with purplish mottling, but occasionally pale tan. They lacked the central scar so common in papillary carcinoma. Most frequently they were circumscribed though unencapsulated (Fig. 1), but in 1 case the tumor involved the thyroid more generally (Fig. 2). There were no multicentric tumors. The neoplasms occurred in thyroids that revealed no nodular change as well as in nodular goiters (8 cases). The tumors were generally of moderate size, varying in their greatest diameter from 1.5 to 8.0 cm. With the accompanying thyroid tissue, they weighed from 12 to 140 Gm. Usually, the thyroid capsule was intact; though its adherence to adjoining structures was increased, it was not directly penetrated by the tumor. In 2 cases, gross neoplasm was evident in the thyroidal capsular veins. In 1 case of extensive thyroidal and lymph node involvement in which a mediastinal dissection was performed, there was invasion of the innominate vein from an adjoining metastatic focus. Cervical lymph node metastasis was evident in 10 patients (48 per cent) at the time of initial operation. Mediastinal lymph node involvement was demonstrated in 1 case in which operative dissection was performed, and mediastinal masses were roentgenographically evident during the course of the disease in 2 instances. The capsules of the involved lymph nodes were more often intact than violated, but the degree of adherence of lymph nodes to adjoining

FIG. 1. Circumscribed neoplasm in thyroid lobe. The tissue was gray-white with patchy reddish-purple mottling.

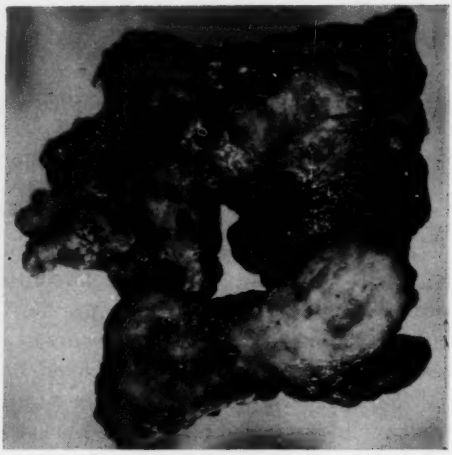


FIG. 2. Neoplasm replacing most of thyroid; massive metastases in adjoining tissue (radical neck dissection).

structures usually was greater than that observed with papillary carcinoma In 2 patients, infiltrating masses were present in the areas of lymph node involvement.

Microscopically the basic pattern was one of sheetlike growth, the neoplastic masses being interrupted by an irregular trabecular stroma of variable density which served to separate the neoplasm into fairly discrete groups of cells and islands of variable size and contour (Figs. 3 and 4). A palisaded arrangement of marginal cells in tumor islands occasionally was observed. Stromal amyloid was present in all tumors in the series (Figs. 4 and 5). It often was evident in the routine sections stained with hematoxylin-eosin, appearing as a bright red or pink, granular or homogeneous substance. The crystal violet stain was used for confirmation of the presence of amyloid in all tumors, and in a few instances was necessary for its recognition. Occasionally there was a foreign body giant-cell reaction about the amyloid patches. The neoplastic cells were of medium size (from 9 to 16 microns in greatest diameter) and of spindle, polyhedral, angular or round shape. In one neoplasm the cells were of such elongated spindle type as to resemble those of a spindle-cell sarcoma. The nuclear-cytoplasmic ratio was generally low. The nuclei often were oval or round and similar in size and in staining property. A rare giant nuclear form was present in one neoplasm. Mitoses, though often present, generally were sparse. Neither

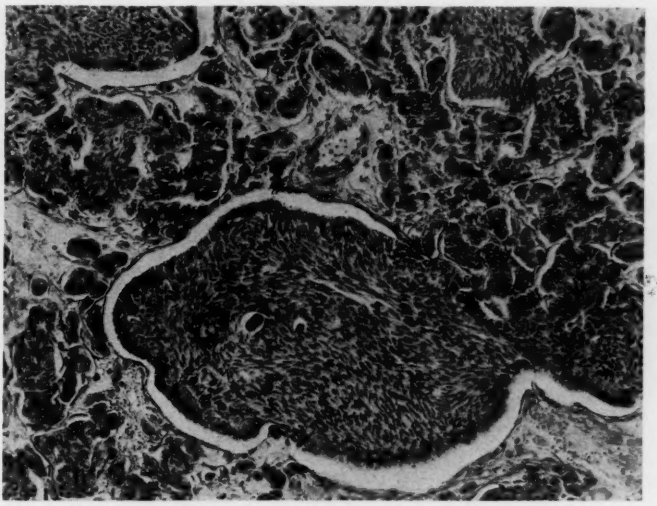


FIG. 3

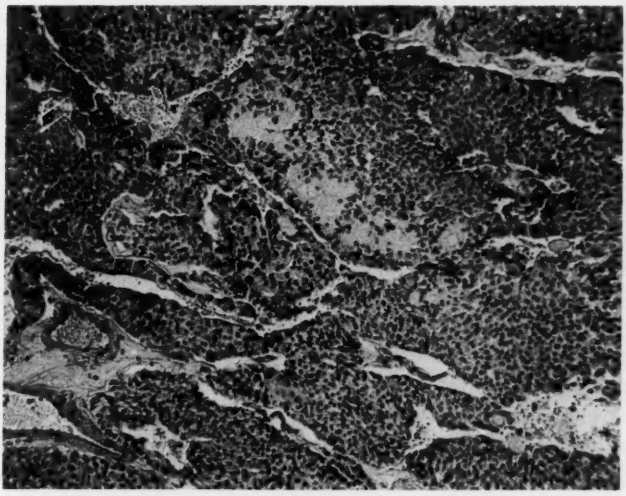


FIG. 4

FIG. 3. Solid sheets of spindle-shaped cells with marginal palisading and irregular smaller groups separated by loose stroma. Hematoxylin-eosin and methylene-blue. Magnification 100×.

FIG. 4. Sheets of tumor cells with narrow bands and homogeneous patches of stroma. The latter stained positively for amyloid with crystal violet. Hematoxylin and eosin. Magnification 100×.

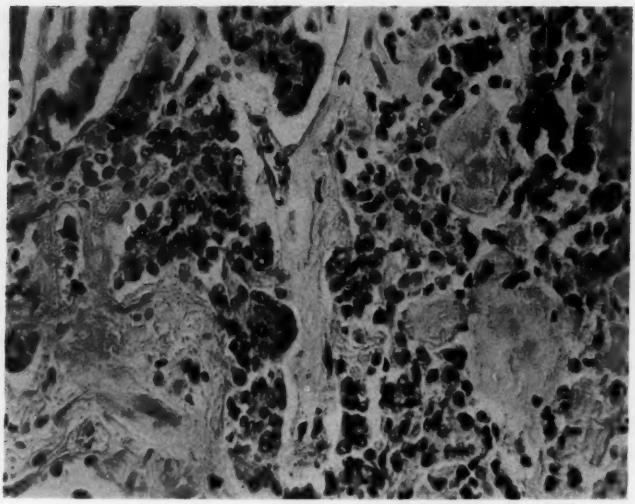


FIG. 5. Irregular groups of neoplastic cells with patches of dense stroma. The latter areas showed a positive reaction for amyloid with crystal violet. Hematoxylin-eosin and methylene blue. Magnification 320×.

neoplastic giant cells nor atypical mitoses were evident. In the older formalin-fixed material particularly, the neoplastic cells were slightly separated from one another and from the stroma, producing irregular spaces but not forming a specific structure. Occasionally such loosely arranged areas presented a pseudopapilliferous pattern as the individually detached cells separated from those still covering patches of stroma. Psammoma bodies were not observed. The solid arrangement of predominantly polyhedral cells in several tumors was suggestive of the structure of carcinoid (argentaffinoma), but silver stains did not reveal any granules. One tumor exhibited festoons of anastomosing cords of cells about delicate stromal vessels and thus simulated islet-cell tumor. In 3 neoplasms, in addition to the predominantly basic solid pattern, there were sparse follicular elements, some of which appeared to represent thyroid follicles included in the invading tumor. Though not sharply demarcated from the thyroid tissue, neoplastic elements often infiltrated the adjoining gland en masse rather than as individual lesions.

CLINICAL FEATURES

Fourteen of the tumors occurred in women, and 7 in men. The ages of the patients ranged from 33 to 66 years, the median age being 50 years. There were no particularly favored locations as to thyroid involvement; the majority appeared to be in one or other of the lateral lobes.

Metastases were present in the cervical lymph nodes in 10 of 21 pa-

tients (48 per cent) at the time of the initial operation; in 2 patients cervical lymph node metastasis developed two and three years, respectively, following the thyroid surgery. Thus in a total of 12 patients (58 per cent), secondary carcinoma developed in cervical lymph node chains.

Surgical treatment was individualized and variable, depending upon the extent of the lesion; it consisted of either lobectomy or subtotal thyroidectomy (about equally divided in the series) with or without lymph node dissection or radical neck dissection. Eleven of the 21 patients died; in 6, death occurred because of neoplasm, in four months, and in one, seven, nine, sixteen and twenty-seven years respectively after the initial thyroid operation. Twelve patients were known to have survived for more than five years after the initial thyroid surgery, namely, for six, seven, seven, seven, eight, eight, nine, thirteen, fourteen, sixteen, twenty-five and twenty-seven years (Fig. 6). One patient survived for fourteen years after cervical lymph node metastasis developed, first five months and then two years after the initial treatment. Recurrences may take place many years following the initial thyroid operation; in our series there was recurrence in a surgical scar after four years, as a superior mediastinal mass after nine years, and in the remaining thyroid tissue after fourteen years. Cervical lymph node recurrences were observed in 5 cases; 4 of these patients (2 of whom had undergone radical neck operations initially) died of their disease seven, nine, sixteen and twenty-seven years respectively after definitive surgical treatment. Cervical lymph node metastasis was present in 5 of 8 patients in the group surviving 0-through-5 years, in 1 patient of the group surviving 5-through-10 years, and in 4 of 5 patients in the group surviving more than 10 years. In patients who died because of the neoplastic disease, neoplasm was manifested as a distant metastasis (lung, 2 cases; liver, 2 cases; bone, 3 cases) and death was not directly due to the local recurrence.

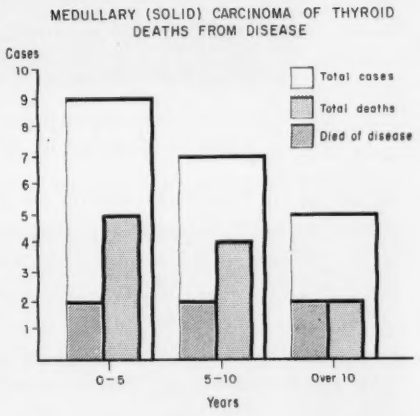


FIGURE 6

DIFFERENTIAL DIAGNOSIS

Clinically, because of the lymph node metastasis, these tumors are liable to be regarded initially as papillary carcinoma. Histologically, the solid and nonfollicular pattern may result in interpretation of the neo-

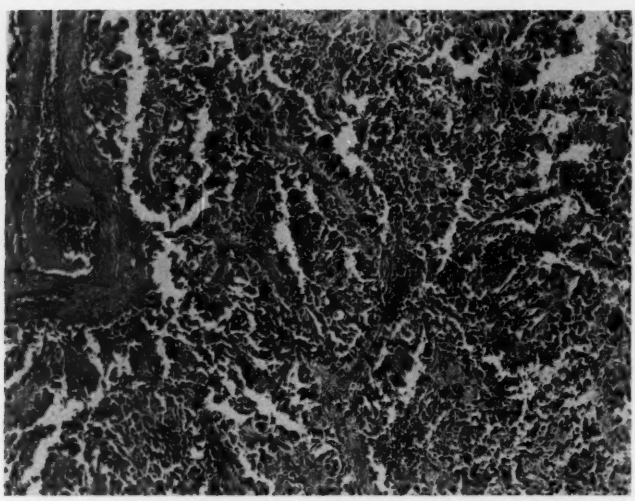


FIG. 7. Artifactual separation of tumor cells producing pseudopapilliferous pattern. Hematoxylin and eosin. Magnification 80×.

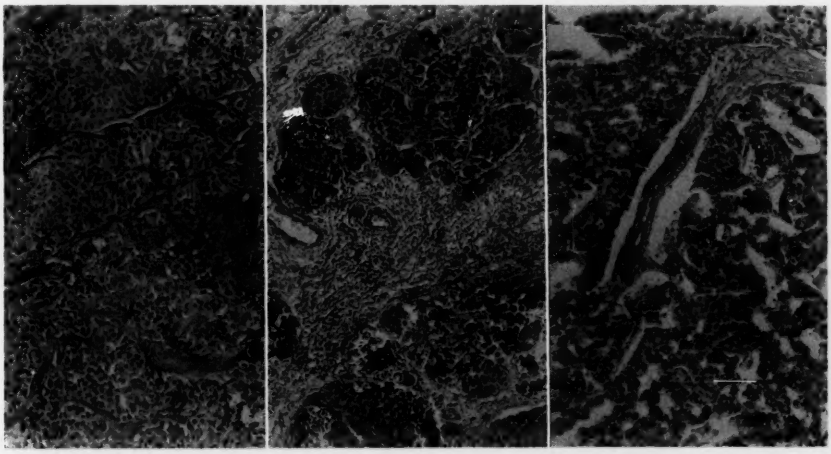
plasm as an "undifferentiated carcinoma," thus raising the possibility of confusion with the highly malignant anaplastic carcinoma and assigning to the tumor under examination a much more serious potentiality than it appears to possess. The medullary neoplasm lacks the anaplasia of tumors of the highly malignant group, exhibits less frequent mitoses, and does not contain areas of necrosis or polymorphonuclear infiltration. It is undifferentiated in the sense that the dominant pattern of growth does not include the production of follicles, but it is far different clinically and pathologically from the anaplastic type of undifferentiated tumor. Because of the pseudopapilliferous pattern, especially in the old formalin-fixed tissues (Fig. 7), the possibility of a basically papillary carcinoma may be a consideration, although precisely formed papillary structures are not a feature.

The neoplasm in some instances may histologically mimic other neoplasms, and this is of importance in considering metastasis. The structure of angular, spindle-like cells sometimes approximates the appearance of transitional-cell carcinoma of the bladder (Fig. 8*a*), but the stroma usually is a more prominent feature and is more dense in the thyroid tumor. When polyhedral and round-cell forms are predominant, the neoplasm may bear a resemblance to a carcinoid tumor (argentaffinoma) (Fig. 8*b*). In 1 instance the neoplasm resembled an islet-cell tumor (Fig. 8*c*). The distinctive features of medullary carcinoma are the solid histologic pattern of an invasive tumor, relatively uniform cells, and stromal amyloid. Usually the amyloid is evident in routine sections, but in a few tumors the crystal violet

stain is necessary for its demonstration. The presence of amyloid in the stroma is of special significance in properly assessing the origin of a neoplasm when interpreting a metastatic lesion. Plasmocytoma is the only other tumor having such a common association with amyloid, but this tumor would cause no histocytologic confusion in diagnosis.

DISCUSSION

Considerable thought has been given to the selection of a term that will indicate the distinctive features of the histologic pattern of this tumor. Descriptively, it is solid and invasive but it is important to distinguish it from other thyroid carcinomas with a sheetlike pattern. "Solid" carcinoma was considered as a definitive term, but since this is one of the growth patterns seen in anaplastic carcinoma, also an undifferentiated type, it was not regarded as an entirely desirable specific designation according to present understanding, without qualification. An objection to the use of the term "medullary" may be made on the basis that a tumor so designated should be of soft character, grossly. However, this is also the most commonly used term for a histologically solid type of tumor, often of intermediate malignancy. It therefore seemed reasonable to employ this designation, since it serves as a distinctive term, indicates properly the histologic



a b c

FIG. 8. Patterns of medullary carcinoma simulating other tumors. (**a**) Solid masses of spindle-shaped and polyhedral cells with narrow, vascular stromal bands, simulating transitional-cell carcinoma of the bladder. Hematoxylin and eosin; magnification 80×. (**b**) Irregular nests of polyhedral cells separated by fibrous stroma, simulating carcinoid. Magnification 100×. (**c**) Festoon-like arrangement of tumor cells with intervening stroma and vessels, simulating islet-cell tumor. Magnification 80×.

pattern of the tumor, is more specific than the term solid carcinoma, and has only the disadvantage of improperly indicating the gross consistency of the tumor.

In most previous reports, it has been difficult to locate the exact assignment of these neoplasms within the classification of types of thyroid tumor. In 1953 we and our associates (1) made a brief review of 12 cases of this carcinoma as part of a presentation on neoplasms of the thyroid, and concluded that medullary carcinoma belonged to a group of neoplasms of intermediate malignancy. Horn (2) apparently included some of these tumors in his description of thyroid carcinomas of unusual configuration and type. These carcinomas, however, were not separated from the other tumors described in his report which were more malignant and aggressive anaplastic carcinomas. He was impressed by the peculiar tendency to lymph node metastasis but it may be noted that anaplastic carcinoma also has this potentiality.

Brandenburg (3), in a case report, described as a "metastasizing amyloid struma" a thyroid neoplasm which appears to belong to the category of medullary or "solid" carcinoma. The patient was a 32-year-old man with a ten-year history of goiter. The thyroid lesion was diagnosed as a malignant struma. Histologically, the neoplasm and subsequent lymph node metastasis revealed patches of amyloid in the stroma and a pattern that appeared similar to that in the tumors of the present series. Brandenburg suggested that the amyloid may be elaborated by the neoplastic cells, and represent an altered form of colloid. He also cited another case published by Stoffel (4) which may be in this category.

The presence of amyloid in the stroma appears to be practically distinctive for this neoplasm among those of thyroid origin. Indeed, except for plasmocytoma, there is no other tumor with such a predilection for amyloid deposition. Other carcinomas of the thyroid (including papillary and anaplastic types) studied after hematoxylin-eosin staining showed no suggestion of the patchy bright-pink or red-staining material found in the stroma of these neoplasms, nor did metachromatic staining for amyloid reveal the presence of this substance in 15 thyroid carcinomas selected because of unusually pink-staining stroma. Whether amyloid is a constant component of the stroma in this particular type of tumor cannot be stated on the basis of this relatively small series. That it may vary in amount is demonstrated by the fact that reactions to crystal violet were weak in 2 of the 21 tumors. In a complete necropsy of a patient who died of a medullary carcinoma of the thyroid with massive neck and mediastinal metastases, no amyloid was demonstrable in tissues other than the neoplasm. Furthermore, there was no sign of amyloid disease in any case in this series.

Serum paper electrophoresis determinations, available in 1 instance, did not show any abnormality of the protein fractions.

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PARENCHYMAL FINDINGS IN THYROIDAL CARCINOMA: PATHOLOGIC STUDY OF 256 CASES*†

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ABSTRACT

In this study an attempt is made to describe the type of thyroid gland in which cancer is found. Sixty-five per cent of the 256 cancers studied occurred in otherwise non-nodular glands. The cancers were grouped in 5 categories: papillary, follicular, solid and anaplastic carcinomas and malignant lymphomas. Eight glands contained malignant lymphomas. About 90 per cent of the carcinomas in the series occurred in glands with no or minimal lymphocytic infiltration of the parenchyma. The incidence of struma lymphomatosa was less than 1 per cent and the incidence of moderate to marked lymphocytic infiltration was about 10 per cent. Nodularity and lymphocytic infiltration occurred in the portion of the glands exclusive of the cancers with about the same frequency as in glands obtained at necropsy and reported in other papers. Most of the 256 cancers occurred in otherwise essentially normal glands. Lymph-node metastasis was found to occur more frequently from those papillary carcinomas that were found in glands in which the parenchyma was otherwise non-nodular and free of lymphocytic infiltration than from similar carcinomas that were found in glands in which the parenchyma was nodular or infiltrated by lymphocytes.

UNTIL about ten years ago most writers on the subject expressed the opinion that nearly all carcinomas of the thyroid gland arose in or from pre-existing benign neoplasms (1–9). In 1955 Dailey, Lindsay and Skahen (10) reported that they had reviewed cases in which thyroid carcinomas arose in glands involved by Hashimoto's disease (chronic thyroiditis) more frequently than would be expected on the basis of chance. This

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† Abridgment of thesis submitted by Dr. Meier to the Faculty of the Graduate School of the University of Minnesota in partial fulfillment of the requirements for the degree of Master of Science in Surgery.

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relationship was especially interesting to us and was an important reason for initiation of the present study.

MATERIALS AND METHODS

Cases in which a diagnosis of cancer of the thyroid gland was made at the Mayo Clinic during the five-year period, 1951 to 1955 inclusive, were reviewed and 256 were selected for study. The principal criteria for selection were 1) a primary histologic diagnosis of either carcinoma or malignant lymphoma of the thyroid gland, and 2) an adequate amount of thyroid parenchyma for study. We have used the word "parenchyma" to indicate thyroid tissue exclusive of malignant disease and the word "cancer" to indicate both carcinomas and malignant lymphomas.

The cancers were grouped into the following 5 categories: papillary, follicular, solid and anaplastic carcinomas and malignant lymphomas. Tumors composed of mixed papillary and follicular structures in any proportion were classified as papillary. Whenever the condition permitted, the surgeons performed total lobectomy on the side involved by the cancer and fairly extensive subtotal lobectomy on the other side. In 128 cases, less than 3 or 4 mm. of parenchyma was adjacent to the carcinoma; these cases were not included in the study because of inadequate specimens of parenchyma. Of these 128 cases, 65 were classified as papillary, 25 as follicular, 14 as solid and 20 as anaplastic. A greater number of cases was excluded from the group having anaplastic lesions, since the operation performed in many instances was necessarily limited in scope.

Gross specimens were reviewed individually. Blocks were cut from the parenchyma of the glands, from the malignant portions, and from metastatic lesions. Paraffin sections stained with hematoxylin and eosin were prepared from these blocks and were employed in classification of the tumors and parenchyma.

The parenchyma of each gland was categorized as to nodularity and degree of lymphocytic infiltration. Changes of granulomatous thyroiditis and of parenchymatous hypertrophy and hyperplasia also were sought for and noted.

RESULTS

Nodularity of the parenchyma. Nodularity of the parenchyma was present in about 35 per cent of the 256 cases and "true" adenomas were present in 4.3 per cent of the glands (Fig. 1). The remaining glands, which represented about 65 per cent of the 256 cases, were, with the exception of the cancer, free of nodules. The nodules were generally of the type considered to result from cycles of hypertrophy and involution, were composed of large follicles, and were not clearly encapsulated. Their structure did not vary greatly from the surrounding tissue but often contained cysts and calcified or fibrotic regions. The number of nodules varied from 1 to 20 or more per gland and they were more than 5 mm. in diameter. The so-called true adenomas were encapsulated, were composed of tissue distinctly different from the surrounding parenchyma (generally of fetal thyroid tissue), and were free of lobulation (11). For the most part the diagnosis of nodularity was based on examination of gross specimens.

There were more than two and a half times as many females as males in

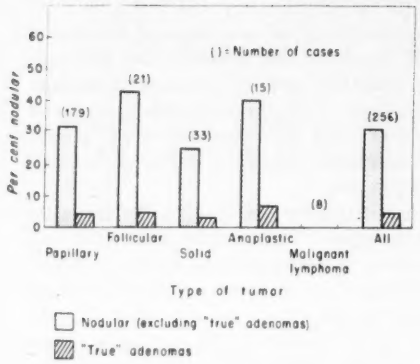


FIG. 1. Percentage of cases in each cancer group with nodular parenchyma.

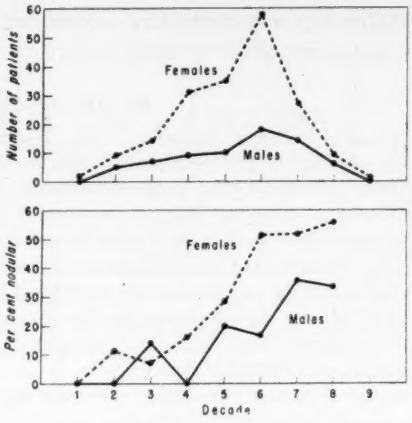


FIG. 2. Number of cases by decades and percentage of cases in each decade with nodular parenchyma ("true" adenomas excluded).

the series and in both sexes the greatest number of patients was in the sixth decade of life (Fig. 2). The lower graph is a plot of the incidence of parenchymal nodularity versus age by decades. It is apparent that the incidence of nodularity increases with the patient's age and that nodularity is more frequent among female patients. These same trends have been noted in necropsy series (12–15).

It is of interest that the peak occurrence of carcinoma and the sharp increase in incidence of nodularity of the parenchyma occur in the same age group. Many patients with nodularity of the parenchyma probably came to operation because of this nodularity and not because of signs or symptoms due to cancer. The increase in the number of cases of cancer among patients in the fifth and sixth decades of life may reflect, in part, the increased incidence of parenchymal nodularity in these age groups; it also suggests, however, that related factors may account for both the cancer and the parenchymal nodularity. This does not necessarily indicate a cause-and-effect relationship.

Lymphocytic infiltration of the parenchyma. Figure 3 presents the degree of lymphocytic infiltration found in the parenchyma of the glands studied. The columns on the left in each category represent the percentage of patients with no more than a trace of lymphocytic infiltration. Fifty-eight per cent of the glands had only a trace of infiltration whereas 29 per cent had minimal infiltration. A smaller proportion (11.7 per cent) had a moderate to marked degree of lymphocytic infiltration, and slightly less than

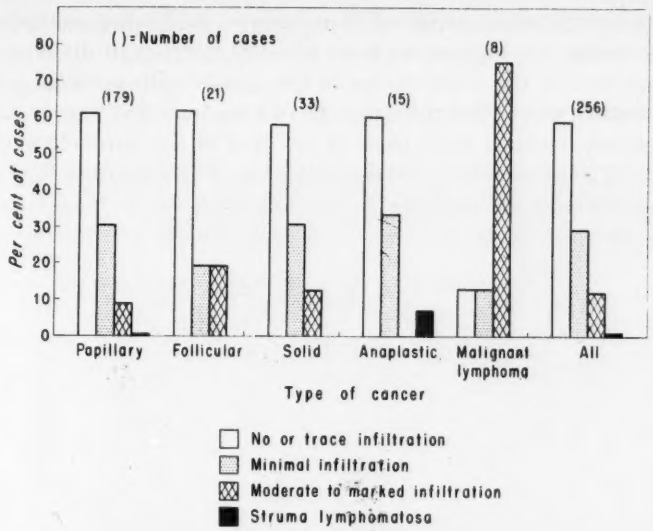
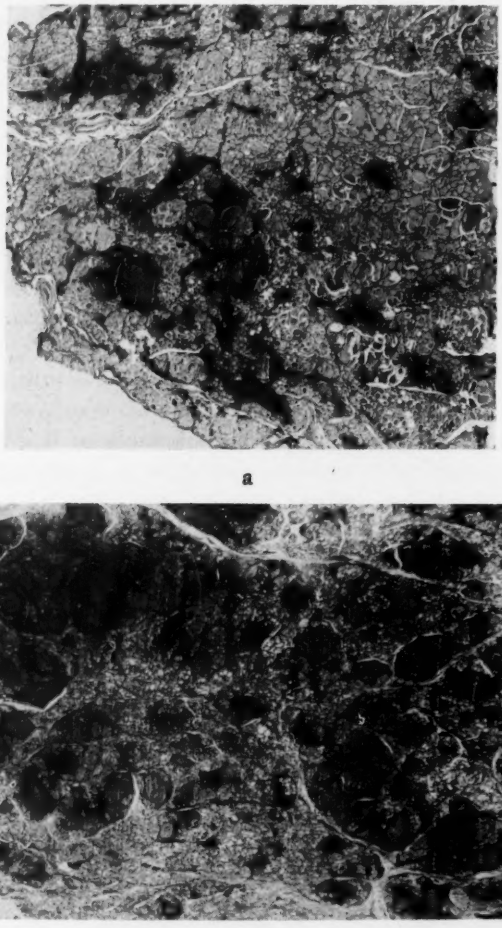


FIG. 3. Percentage of cases in each cancer group with various degrees of lymphocytic infiltration of the parenchyma.

10 per cent of the glands with carcinomas had this degree of infiltration. Two carcinomas in the series were found in glands which were involved by generalized changes of typical struma lymphomatosa. It is worthy of note that 6 of the 8 malignant lymphomas occurred in glands with moderate-to-marked lymphocytic infiltration.

FIG. 4. Parenchyma of thyroid with minimal lymphocytic infiltration. The dark portions are infiltrated by lymphocytes (hematoxylin and eosin; ×8).

The glands with only a trace of lymphocytic infiltration contained, at most, one clump of lymphocytes less than 100 microns in diameter. Less than 10 per cent of the parenchyma of the glands with minimal lymphocytic infiltration was replaced by clumps of lymphocytes (Fig. 4). Clumps of lymphocytes replaced more than 10 per cent of the parenchyma in the glands having moderate-to-marked infiltration (Fig. 5*a* and *b*), but diffuse changes associated with typical struma lymphomatosa were lacking. Four of these glands, however, did have localized changes of struma lympho-



a

b

FIG. 5. **a.** Moderate lymphocytic infiltration of parenchyma of thyroid (hematoxylin and eosin; ×8). **b.** Marked lymphocytic infiltration of parenchyma of thyroid (hematoxylin and eosin; ×8).

matosa. Hellwig found the incidence of lymphocytic infiltration in thyroid glands obtained at necropsy to be 16 per cent (16) and Rice reported an incidence of 10 per cent (14).

Two glands were involved by diffuse changes of typical struma lympho-

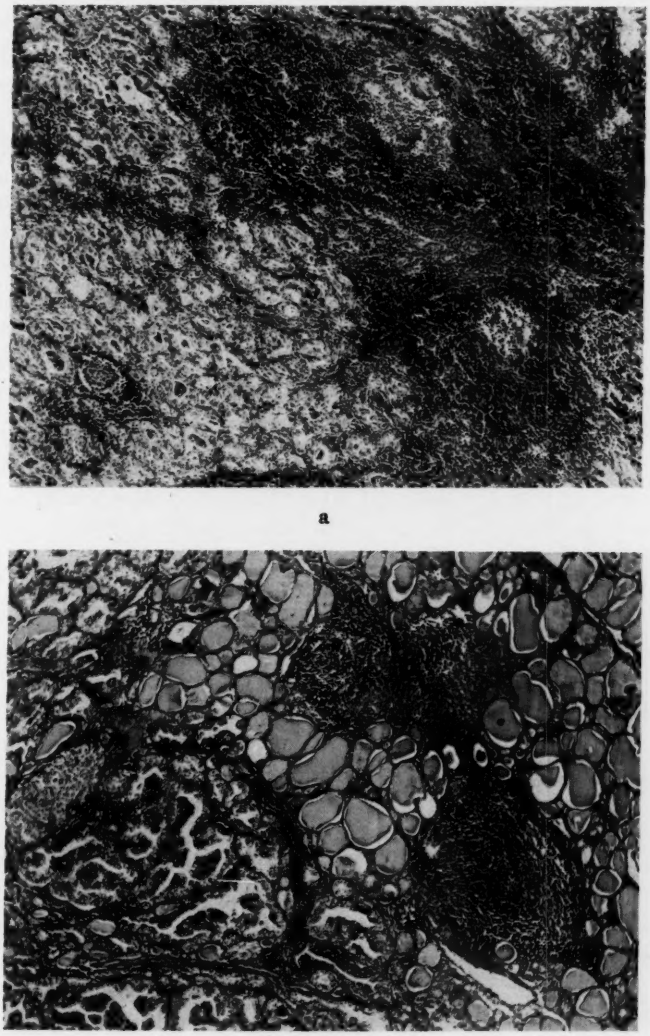


FIG. 6. **a.** Parenchyma of thyroid involved by struma lymphomatosa (hematoxylin and eosin; ×60). **b.** Papillary carcinoma with lymphocytic infiltration adjacent to the carcinoma (hematoxylin and eosin; ×60).

matosa (Fig. 6*a*). The epithelial cells contained abundant acidophilic cytoplasm; there was a marked degree of lymphocytic infiltration, and lymphocytes and plasma cells infiltrated the interfollicular spaces (17, 18).

The areas adjacent to the carcinoma were infiltrated by lymphocytes in more than a third of the glands containing papillary carcinomas (Fig. 6*b*). Infiltration in the immediate vicinity of the carcinoma was considered to be a reaction to the carcinoma and was ignored in the classification of degree of infiltration of the parenchyma if specimens taken at a greater distance from the carcinoma were free of infiltration.

Minimal infiltration of the parenchyma occurred with greater frequency in the glands of females and was found with greatest frequency in patients in the fifth and sixth decades (Fig. 7). Moderate-to-marked infiltration also occurred much more frequently in the glands of females but the incidence varied less with age.

Relation of nodal metastasis to parenchymal changes. Among the cases of papillary carcinomas, nodal metastasis was most frequent in those patients whose glands were most nearly normal. As a rule, regional lymph nodes were removed when they were palpable clinically or at operation, so that the figures for nodal metastasis are comparable although not absolute.

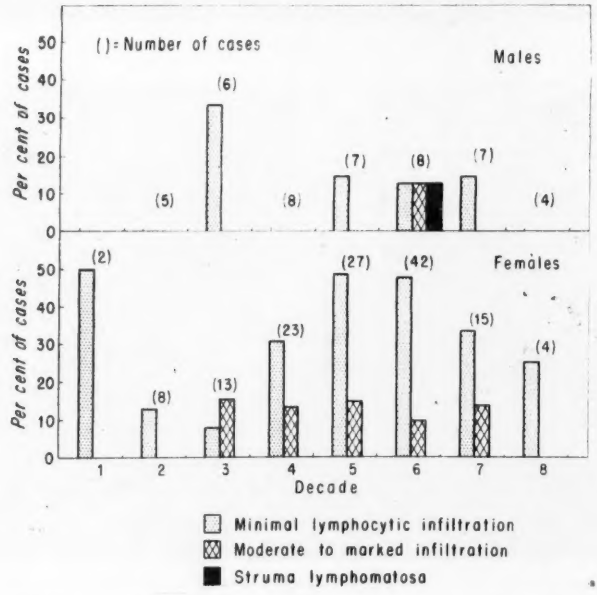


FIG. 7. Incidence of lymphocytic infiltration of the parenchyma by decades for group with primary diagnosis of papillary carcinoma.

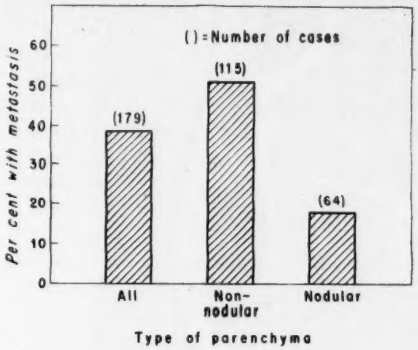


FIG. 8. Incidence of lymph node metastasis from papillary carcinoma occurring in nodular and non-nodular parenchyma.

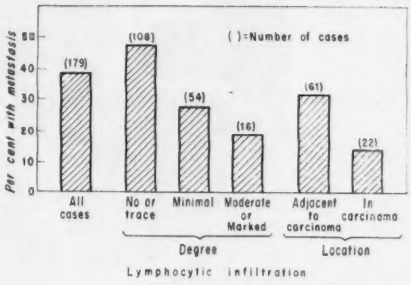


FIG. 9. Incidence of lymph node metastasis from papillary carcinoma occurring in parenchyma with different degrees of lymphocytic infiltration.

Figure 8 shows the incidence of regional metastasis to lymph nodes for papillary carcinomas occurring in glands with nodular and non-nodular parenchyma. The surprising thing is that the incidence of nodal metastasis is almost three times as great in the group with non-nodular parenchyma. It seems unlikely that nodularity of the parenchyma protects against metastasis; more than likely many of the carcinomas occurring in nodular parenchyma were brought to the attention of the patient or the physician by the nodularity, whereas many of the carcinomas occurring in non-nodular parenchyma were brought to light by signs or symptoms caused by metastasis. Almost 39 per cent of the papillary carcinomas that occurred in nodular parenchyma were less than 1 cm. in diameter, and about 27 per cent of the papillary carcinomas occurring in non-nodular parenchyma were diagnosed by biopsy of lymph nodes.

Nodal metastasis was found in 39 per cent of the 179 patients with papillary carcinomas (Fig. 9). The group with no more than a trace of lymphocytic infiltration in the parenchyma had an incidence of metastasis to the lymph nodes of 47 per cent. The group with minimal infiltration of the parenchyma had an incidence of metastasis of only 28 per cent and less than 20 per cent of the patients with moderate-to-marked parenchymal infiltration had nodal metastasis. Metastasis occurred with less frequency among the patients having lymphocytic infiltration adjacent to the carcinoma than it did among the papillary group of patients as a whole. The cases in which there was a great amount of lymphocytic or plasma-cell infiltration of the carcinoma itself had the lowest incidence of metastasis; such infiltration (Fig. 10) was found in 22 cases, 3 of which had metastasis.

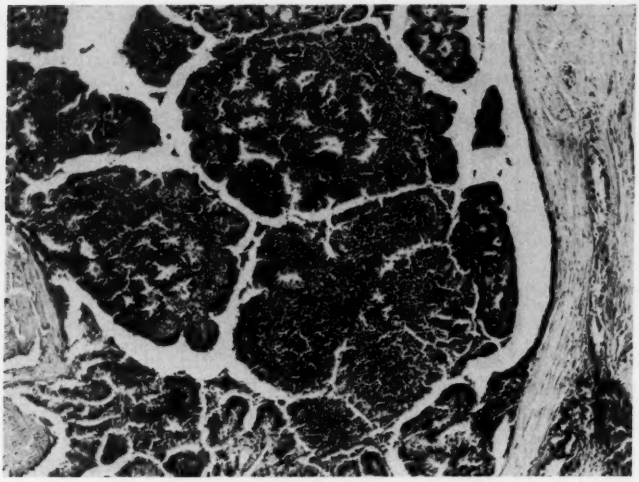


FIG. 10. Papillary carcinoma with infiltration of the carcinoma by numerous lymphocytes and plasma cells (hematoxylin and eosin; ×60).

Miscellaneous changes in parenchyma. In 3 glands, small areas of granulomatous thyroiditis had developed in the tissue external to the cancer (19). These changes had not caused symptoms in any of these cases. Histologic changes generally associated with exophthalmic goiter were found in the parenchyma of 4 glands. Symptoms and signs of hyperthyroidism were present in 3 patients in whom the parenchyma of the gland showed these changes.

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The Endocrine Society

THE 1959 ANNUAL MEETING

The Forty-First Annual Meeting of The Endocrine Society will be held in the Haddon Hall Hotel, Atlantic City, New Jersey, Thursday, Friday, and Saturday, *June 4, 5 and 6, 1959.*

The Chairman of Local Arrangements is Dr. Matthew Molitch.

Scientific Sessions will be held from 9:00 A.M. to 5:00 P.M. daily, and in addition there will be simultaneous afternoon sessions. The annual dinner is scheduled for Friday, June 5, at 7:30 P.M. preceded by cocktails at 6:30 P.M.

All members are urged to make their hotel reservations immediately. The Chalfonte-Haddon Hall Hotel will hold 300 bedrooms for members until May 1, 1959, after which time the hotel will not guarantee further reservations. Therefore it is imperative that you make your reservations now directly with the hotel, advising them of time and date of arrival and departure. Avoid disappointment.

Final program, membership card and advance registration forms will be sent on May 1, 1959 to members whose current dues have been paid.

Those wishing to present papers, which will be strictly limited to ten minutes, should send four copies of the title and abstract to the Vice-President, Dr. Frank Engel, Duke Hospital, Durham, North Carolina, not later than *February 1, 1959. It is imperative that the abstracts be informative and complete with results and conclusions*—not a statement that those will be presented at the meeting—in order that they may be of reference value and suitable for printing in the program. The reading and processing of approximately two hundred abstracts submitted each year and the compiling of the program from these involves a tremendous amount of time and effort on the part of the Program Committee. The Council requests that the authors adhere strictly to the following rules when submitting abstracts, otherwise they cannot be considered:

1. *It is assumed that abstracts submitted for this program have not and will not be submitted elsewhere.*
2. Abstracts may not exceed two hundred words, or equivalent space, exclusive of title. No footnotes or acknowledgments to sponsors can be published. References, if used, must be placed in the body of the

text. The abstract should consist of a single paragraph, if possible. Structural chemical formulas cannot be used.

3. The title heading must be arranged as follows:
   Line 1. Title, not to exceed fifteen words.
   Line 2. Author/s. The name of each nonmember-author collaborating with member-authors is to be followed by the phrase "(by invitation)." Names of the nonmembers who are introduced, *i.e.*, who are not collaborators with member-authors, are to be followed by the phrase "(introduced by . . .)." The principal degree, *e.g.*, M.D., of each author should be written after his name.
   Line 3. Institution of origin and city in which institution is located.
4. The body of the abstract, typed double-space, should follow the heading. The original copy should be on bond paper. There should be three copies.
5. Abstracts should be letter perfect, since there will be no opportunity for proof reading by the authors.

THE 1959 AWARDS

The selection of the recipients of the awards of The Endocrine Society is made by a Committee appointed by Council of the Society. These awards and fellowships carry no obligation by the recipient to the Society or to the donors.

FRED CONRAD KOCH AWARD

During the past year a substantial legacy has been bequeathed to the Society by the late Elizabeth Koch for the purpose of establishing the Fred Conrad Koch Memorial Fund in memory of her late husband, Distinguished Service Professor of Physiological Chemistry at the University of Chicago and pioneer in the isolation of the androgens. This is to be the highest honor of The Endocrine Society and is to be represented by a medal that is to be known as the Koch Medal of The Endocrine Society. The medal, as well as an honorarium of $3,500, is to be given annually to an individual for work of special distinction in endocrinology. The recipient shall be chosen from nominations presented by members of the Society and is limited to citizens of the United States and Canada.

This Award will replace the Medal of The Endocrine Society which was established in 1954 and presented to Dr. Carl R. Moore in 1955, Dr. Frederick L. Hisaw in 1956, Dr. Joseph C. Aub in 1957 and Dr. I. L. Chaikoff in 1958. The Medal of the Endocrine Society replaced the E. R. Squibb Award which was formerly the highest honor bestowed by the Society.

Past recipients of the Squibb Award were Dr. George W. Corner in 1940, Dr. Philip E. Smith in 1941, Dr. Fred C. Koch in 1942, (no award in 1943), Dr. E. A. Doisy in 1944, Dr. E. C. Kendall in 1945, Dr. Carl G. Hartman in 1946, Drs. Carl F. and Gerty T. Cori in 1947, Dr. Fuller Albright in 1948, Dr. Herbert M. Evans in 1949, Dr. C. N. H. Long in 1950, Dr. J. B. Collip in 1951, Dr. James H. Means in 1952 and Dr. David Marine in 1953.

THE CIBA AWARD

The Ciba Award, to recognize the meritorious accomplishments of an investigator not more than 35 years of age in the field of clinical or preclinical endocrinology, was established in 1942, but no recipient was selected in 1942 or 1943. In 1944 the Award was presented to Dr. E. B. Astwood; 1945—Dr. Jane A. Russell; 1946—Dr. Martin M. Hoffman; 1947—Dr. Choh Hao Li; 1948—Dr. Carl Heller; 1949—Dr. George Sayers; 1950—Dr. Oscar M. Hechter; 1951—Dr. Albert Segaloff; 1952—Dr. Seymour Lieberman; 1953—Dr. Sidney Roberts and Dr. Clara Szego (Mrs. Roberts); 1954—Dr. Isadore M. Rosenberg; 1955—Dr. Jack Gross; 1956—Dr. Alfred M. Bongiovanni; 1957—Dr. Nicholas S. Halmi; 1958—Dr. Monte A. Greer. Prior to 1952 the Award was $1,200. It has now been increased to $2,500.

THE AYERST AND THE SQUIBB FELLOWSHIPS

The Ayerst Fellowship was established in 1947 and the Squibb Fellowship in 1956. They are designed to assist men or women of exceptional promise in furthering their advancement towards a career in endocrinology. Each Fellowship is awarded on alternate years and the stipend, which will not exceed $5,000, may be divided into two Fellowships in varying amounts in accordance with the qualifications of the appointees. Individuals possessing the M.D. or Ph.D. degree, or candidates for either of these degrees, are eligible for appointment.

Applicants must submit the following information:

1. Evidence of scientific ability as attested by studies completed or in progress.
2. Recommendations from individuals familiar with the candidate and his work.
3. A proposed program of study.
4. Acceptance of the individual by the head of the department in which the Fellowship will be held.
5. A statement that he or she will serve full time if awarded a Fellowship. A small amount of time (10 to 15 per cent) may be spent in course work or participation in teaching, the latter purely on a voluntary basis.

THE SCHERING AND THE UPJOHN SCHOLARS OF THE ENDOCRINE SOCIETY

The Council of The Endocrine Society has established a category of Scholars. These grants have been made available through the generosity of the Schering Corporation and the Upjohn Company, and will be awarded to established investigators and teachers in the field of endocrinology who wish to extend their opportunities for work either in this country or abroad.

The awards will not exceed $2,500 annually for each individual and will be granted on the basis of proposals submitted by the applicant. Such applications should include the estimated financial needs. The funds may be used for travel, maintenance and other expenses.

Nominations

Nominations for the Fred Conrad Koch Award, the Ciba Award, and the Ayerst and the Squibb Fellowships may be made by any member of The Endocrine Society. They should be submitted on forms which may be obtained from the Office of the Secretary, 1200 North Walker Street, Oklahoma City 3, Oklahoma. Completed nominations should be returned to the Secretary not later than *October 15* each year.

Proposals for appointments as Scholars of The Endocrine Society should be made in writing by the individual, and addressed to the Secretary of the Society. They should be submitted by *October 15* each year.

The American Goiter Association

THE 1959 ANNUAL MEETING

The next Annual Meeting of The American Goiter Association will be held in the Drake Hotel, Chicago, Illinois, on *April 30, May 1 and 2, 1959.*

THE VAN METER PRIZE AWARD FOR 1959

The American Goiter Association again offers the Van Meter Prize Award of $300.00 and two honorable mentions for the best essays submitted concerning original work on problems related to the thyroid gland. The award will be made at the annual meeting of the Association which will be held in the Drake Hotel, Chicago, Illinois, April 30, May 1 and 2, 1959.

The competing essays may cover either clinical or research investigations, should not exceed 3,000 words in length and must be presented in English. Duplicate typewritten copies, double spaced, should be sent to the Secretary, Dr. John C. McClintock, 149½ Washington Avenue, Albany 10, New York, not later than *January 15, 1959.* The committee who will review the manuscripts is composed of men well qualified to judge the merits of the competing essays.

A place will be reserved on the program of the annual meeting for the presentation of the winning essay by the author if it is possible for him to attend.

JOHN C. MCCLINTOCK, M.D.
Secretary

(For recipients of 1958 Awards, see September 1958 issue of this Journal.)

NEW YORK UNIVERSITY
POST-GRADUATE MEDICAL SCHOOL

announces the following postgraduate course in

GYNECOLOGICAL ENDOCRINOLOGY

A practical, didactic and clinical presentation with emphasis on the therapeutic management of endocrine disorders in the female, including a discussion of intersex will be offered. Endocrine therapy for menstrual abnormalities and ovulatory defects are discussed. Adrenocortical, thyroid, ovarian and pituitary abnormalities are emphasized. Practical diagnostic tests are discussed and demonstrated, as well as the therapeutic use of the newer progestational steroids.

Five-day, full-time course, *March 30 through April 3, 1959.*

Tuition: $125.00

Given under the direction Dr. Herbert S. Kupperman.

For further information, write to the Office of the Associate Dean, NYU Post-Graduate Medical School, 550 First Avenue, New York 16, N. Y.

BOOK REGISTER

Neurological Basis of Behaviour. Ciba Foundation Symposium, edited by G. E. W. WOLSTENHOLME, M.A., B.Ch. and CECILIA M. O'CONNOR, B.Sc. Contributions are by some of the most prominent figures in the field, and include: Early development of ideas relating the mind with the brain; the behaviour of nerve cells; an appraisal of the effects of diencephalic stimulation of conscious animals in terms of normal behaviour; correlated effects in behaviour and electrical brain activity evoked by stimulation of the reticular system, thalamus and rhinencephalon in the conscious animal; the rôle of the temporal cortex in recall of past experience and interpretation of the present; 'the temporal lobe syndrome' produced by bilateral ablations; some basic mechanisms of the translation of bodily needs into behaviour; neurological basis of responses to stress; the rhinencephalon and behaviour; and the neurological site of action of stilbestrol in eliciting sexual behaviour; 400 pages; 109 illustrations; 1958. Little Brown and Company, Boston, Massachusetts. Price, $9.00.

(*Le*) *Placenta humain. Aspects morphologiques et fonctionnels.* By PROF. JEAN SNOECK, Laboratoire de Gynécologie Expérimentale de l'Université Libre de Bruxelles; Clinique de Gynécologie et d'Obstétrique de l'Hôpital Saint-Pierre, with a foreword by PROF. P. GERARD. This book deals with the subject of the human placenta: Part I, Morphology; and Part II, Physiology. In Part I, chapters are devoted to morphogenesis, microscopic structure, and macroscopic characteristics. In Part II, chapters deal with placental permeability, and endocrine function. All aspects of the subject are treated in considerable detail, including extensive bibliographies; 718 pages; 175 figures; 48 tables; 1958. Masson et Cie, Éditeurs, Paris, France. Price, 7,500 fr.

Polysaccharides in Biology. Transactions of the Third Conference, The Josiah Macy, Jr. Foundation, May 29–31, 1957, Princeton, N.J., edited by GEORG F. SPRINGER, M.D., William Pepper Laboratory of Clinical Medicine and Department of Microbiology, University of Pennsylvania School of Medicine, Philadelphia, Pa. Several prominent participants discuss: Homopolysaccharides and Nucleotides and Saccharide Synthesis; 250 pages; 58 figures; 52 tables; bibliographies; 1958. The Josiah Macy, Jr. Foundation, 16 West 46th Street, New York 36, N.Y. Price $4.75.

Reversicon. A Medical Word Finder. By J. E. SCHMIDT, Ph.B.S., M.D., Litt.D., President of The American Society of Grammatolators and Chairman of the National Association on Standard Medical Vocabulary. In this book, words can be found on the basis of their meaning. Ideas, thoughts or concepts are arranged alphabetically in 25,000 entries. Thus the reversicon (meaning-to-word) is the reverse of the standard lexicon (word-to-meaning). It is useful in finding words unknown to the reader and also in retrieving forgotten and vaguely remembered words; 440 pages; 1958. Charles C Thomas, Publisher, Springfield, Illinois.